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PHYTOPATHOLOGY

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ABSTRACTS OF PAPERS ACCEPTED FOR PRESENTATION AT THE THIRTY-EIGHTH ANNUAL MEETING OF THE SOCIETY, CINCINNATI, OHIO, DECEMBER 28 TO 30, 1946

Development of a Fusarium-wilt-resistant glasshouse tomato variety. ALEXANDER, L. J. The commercial wilt-resistant varieties of tomato now available do not possess sufficient resistance to withstand attacks of Fusarium wilt under Ohio glasshouse conditions. Steam sterilization of the soil gives good results but must be repeated annually. Therefore, it seemed desirable to develop a new variety with sufficient resistance to withstand the disease under Ohio glasshouse conditions. Following publication of the work of Bohn and Tucker, certain of their accessions were artificially inoculated at the fifth or sixth true leaf stage when set out in the growing house. Sufficient resistance was shown to justify the initiation of further breeding work which had as its objective the development of a wilt-resistant variety of the Globe type. Following this trial several crosses and back-crosses of the best accessions were made to Globe. The best accessions from these crosses are in the fifth and sixth generations since the last cross to Globe. These have a fair degree of homogeneity. These lines have been selected specifically for resistance to race 1 because it appears to be the most widely distributed. Race 2 is known to be present in only one glasshouse but probably occurs in a few others. Preliminary yield trials are very promising. Wider yield trials are being planned for the spring crop of 1947.

Effect of streptomycin on higher plants. ANDERSON, H. W., and INEZ NIENOW. Streptomycin sulphate was used in a nutrient solution (Shives) to determine effect on seedlings. After germination the seedling roots were immersed in dilutions of from 5 to 200 units/ml. Concentrations beyond 50 units/ml. were toxic to tomato and radish seedlings. Soybeans were not killed but there was marked stunting of lateral roots. Wheat was not injured at 200 units/ml. The characteristic reduction in lateral root development by Streptomycin was similar to the effect of clavacin on corn, bean, and onion roots in earlier studies. Soaking 8 varieties of seeds 12 hours in 200 units/ml. concentration did not prevent germination but when radish was planted in soil, marked injury (stunting and yellowing) was observed. No reduction in damping-off occurred. Assays of plants grown in Streptomycin solution or in sand cultures watered with the solution showed that Streptomycin is absorbed through the roots. Assays indicated that soybean plants grown in 50 units/ml. solution retained 5–10 units/ml. in expressed sap. Plants grown in 100 units/ml. showed higher assays than those grown in 50-unit concentrations. Soybeans containing 4–5 units/ml. when inoculated with *Xanthomonas phaseoli* var. *sojense* had typical symptoms of bacterial pustule, and the organism was readily reisolated from the lesions.

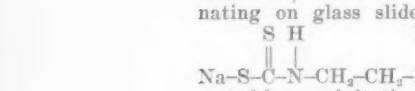
Dutch elm disease recurrence in American elms in relation to the extent of vascular invasion by Ceratostomella ulmi in the year of disease incidence. BANFIELD, W. M. Recurrence of Dutch-elm disease was studied in the season following infection in approximately 700 American elm seedlings that had developed many degrees of vascular invasion as a result of diverse types, locus, and time of infection in the season of initial infection. All trees were in a native stand and varied between 1½ and 4 inches diameter at breast height. The disease did not recur in 276 trees wherein vascular invasion was limited to crown branches. It recurred in 2 of 140 trees wherein vascular invasion was extensive in crown and upper bole. It recurred in 82 of 169 trees wherein vascular invasion was extensive in crown, bole, and roots. Histological examination of these trees revealed vessels of consecutive growth sheaths to be contiguous at numerous points in the root system and in the lower bole. In recovered trees no points were found at which invaded vessels of the sheath of initial infection were in contact with vessels of the new sheath. Invaded vessels of new and old sheaths were contiguous at one or many points in trees in which the disease recurred. Such points of contact were most numerous in the roots, present in the bole, and rare in small branches.

Recurrence of Dutch elm disease in American elms in relation to tree stature. BANFIELD, W. M., E. G. REX, and CURTIS MAY. Wilt and dieback symptoms reappear in the season following infection in American elms as a result of general invasion of the vessels of the new sheath by *Ceratostomella ulmi*. This invasion has its incidence in the infected vessels of the preceding sheath. Trees in which no invasion of new vessels develops from infected vessels of the preceding sheath recover and display no symptoms of disease unless

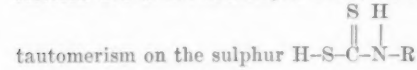
reinfected. Disease recurrence varies with tree stature. The disease recurred in less than 10 per cent of several hundred trees of 1-inch diameter at breast height, which survived the initial year of general vascular invasion. The disease recurred in 49 per cent of 169 trees of 2 to 4.9 inches d.b.h., in 83 per cent of 36 trees of 5 to 9.9 inches, in 92 per cent of 27 trees of 10 to 15 inches, and in all 36 trees, greater than 15 inches d.b.h., on which observations were made. The disease continued to recur each year until death, in all trees but one, wherein it recurred in the second year only. Most trees died in the second season, about a third died in the third season. Three large trees continue as living relicts after 5 years of recurring disease. (Massachusetts State College, N. J. State Department of Agriculture, and U. S. Department of Agriculture.)

Vitamin deficiencies in the Sclerotiniaceae. BARNETT, H. L., and V. G. LILLY. Fifteen species in 8 genera were studied with respect to their growth in the absence and in the presence of 4 common vitamins (thiamin, pyridoxine, inositol, biotin) used separately and in combination. Variations in vitamin requirements were found among the isolates of the same species as well as among different species. Four species (*Sclerotinia sclerotiorum, Botryotinia convoluta, Ciboria pseudotuberosa, Ciborinia erythronii*) were found to be autotrophic with respect to these vitamins. All other species studied were heterotrophic in varying degrees for thiamin or biotin, or both. *Sclerotinia camelliae* and *Lambertella hicoriae* grew well only in a medium containing both thiamin and biotin. Isolates of *Monilinia fructicola, Ciboria acerina,* and *Sclerotinia minor* varied in their response to the vitamins. The growth of certain species was somewhat depressed by pyridoxine or inositol, or both.

Fungicidal mechanism of disodium ethylene bisdithiocarbamate. BARRATT, RAYMOND W., and JAMES G. HORSFALL. Spores of *Macrosporium sarcinaeforme* germinating on glass slides were used to assay the toxicity of numerous derivatives of



Na–S–C(=S)–N–CH_2–CH_2–N–C(=S)–S–Na. The intact molecule may function as previously suggested by precipitating the metals necessary for the enzyme systems of the spore. Lengthening of the aliphatic carbon chain decreased the poteney. The theory is advanced that the major part of the toxicity is due to H_2S liberated during decomposition. The equilibrium Na–S–R + HOH ⇌ H–S–R + NaOH existing in solution provides for hydrogen



tautomerism on the sulphur H–S–C(=S)–N–R ⇌ S=C–N–R, while the hydrogen on the nitrogen

provides for tautomerism between nitrogen and sulphur H–S–C(=S)–N–R ⇌ H–S–C=N–R.

During the dynamic states of the molecule, hydrogen sulphide is evolved: H–S–C(=S)–N–R ⇌ S=C=N–R (ethylene bis diisothiocyanate) + H_2S. Evidence supporting this view is (1) hydrogen sulphide is evolved in water, (2) conversion of the nitrogen from a secondary to a tertiary amine either by methylation or inclusion in a ring reduces toxicity by the order of 20-fold, presumably by preventing resonance, (3) substitution of the sodium by other soluble ions like potassium and quaternary ammonium has no effect on toxicity, but substitution of insolubilizing metallic ions, such as zinc, strontium, or cadmium, reduces toxicity by the order of 2-fold presumably by reducing the rate of hydrolysis.

Cherry yellows and necrotic ring spot of sour cherry in Ontario. No. 1. The value of Prunus persica and P. domestica var. Italian prune as index hosts. BERKELEY, G. H. Necrotic ring-spot symptoms, consisting of delayed foliation, chlorotic spots, rings, and necrosis of leaves, are of the shock type, since affected trees recover, while symptoms of cherry yellows, which consist of yellowing of leaves, which are cast, are of the chronic type in that they recur year after year. Budding experiments indicated that every source of cherry yellows tested has also contained the necrotic ring-spot virus. But necrotic ring spot has been found to occur free from the yellows virus. These experiments indicated that peach seedlings, or the varieties Elberta or Rochester, are of value as indicator hosts. Peaches reacted to necrotic ring-spot virus by causing shock symptoms of delayed foliation, die-back, and bark necrosis, followed by recovery provided the initial shock effect was not too severe. Peaches reacted to the cherry-yellows complex by producing shock symptoms similar to those caused by the necrotic ring-spot virus and a chronic symptom of rosetted shoots which persist. Italian prune reacted somewhat similarly by causing a shock symptom of leaf necrosis with either necrotic ring-spot virus alone, or with the cherry-yellows complex, while the latter in

addition produced chronic symptoms similar to those associated with prune dwarf. However, the experimental evidence does not indicate that cherry yellows and prune dwarf are caused by the same virus or virus complex. Variation in degree of symptoms resulting from inoculation suggests the presence of strains of both viruses.

Alfalfa mosaic on pepper in Ontario. BERKELEY, G. H. An unusual type of mosaic was found on sweet pepper var. California Wonder in Ontario in 1943 and again in 1944. Symptoms included chlorotic rings, spots, and patterns on both leaves and fruit. Preliminary inoculations indicated that the causal virus was closely related to *Marmor medicaginis* H. A large number of inoculations, extending over a 3-year period, using *Marmor medicaginis* H. var. *typicum*, *M. medicaginis* H. var. *solani*, and the pepper virus indicated that the pepper virus is a strain of *Marmor medicaginis*. Comparative studies were made on *Nicotiana tabacum; N. rustica; N. glutinosa; Vicia faba*, broad bean; *Lathyrus odoratus*, sweet pea; *Pisum sativum*, pea; *Phaseolus vulgaris*, bean; *Trifolium pratense*, red clover; *Capsicum frutescens*, pepper; *Soja max*, soybean; *Solanum melongena*, eggplant; *Zinnia elegens*, zinnia; *Cucumis sativus*, cucumber; *Petunia hybrida*, petunia; *Lycopersicon esculentum*, tomato; *Antirrhinum majus*, snapdragon; and *Apium graveolens*, celery. With the following exceptions, all 3 viruses produced similar symptoms on the above hosts. The pepper virus did not infect tomato, while both the alfalfa-mosaic virus and the potato-calico strain did. Whereas the pepper virus and the alfalfa-mosaic virus gave positive results on cucumber, negative results were obtained with the potato-calico virus. The potato-calico strain produced prominent calico symptoms with wide vein-banding on *N. rustica* and *N. glutinosa*. On these same hosts alfalfa-mosaic virus and the pepper virus produced much less prominent symptoms with the wide vein-banding lacking. The pepper virus produced more severe necrosis on *N. glutinosa, N. rustica, N. tabacum* than did alfalfa virus I or the potato-calico virus. Cross protection and thermal death point tests also indicated that the pepper virus was a strain of *Marmor medicaginis* H.

A virus disease of the potato transmitted by the aster leafhopper. BONDE, REINER, and E. S. SCHULTZ. The aster leafhopper, *Macrosteles divisus*, transmitted by artificial inoculations a virus disease of the potato which appears to be different from purple-top wilt. The symptoms of the disease are apparent toward the end of the growing-season and are characterized by a slight dwarfing and by a rolling of the upper leaves of the plant similar to that often associated with rhizoctonia stem rot and other diseases. The stems of the pigmented varieties develop a purplish color. The internodes may become shortened, and the nodes enlarged. The plants do not wilt or die prematurely, and a pronounced necrosis of the vascular bundles near or at the ground line does not occur. None of the tubers from diseased plants becomes flabby or develops vascular discoloration. They germinate normally and the disease is perpetuated through the seed tubers. This disease has been found in potato fields in Aroostook County, Maine, but apparently is of little economic importance. The disease is very similar to apical leafroll, previously described by Schultz and Bonde. (Maine Agricultural Experiment Station and United States Department of Agriculture.)

Necrosis of soybean stem and root caused by Rhizoctonia solani. BOOSALIS, M. G. On stems and roots of soybean attacked by *Rhizoctonia solani* are necrotic lesions in which a browning and discoloration of host cells may extend for 5 to 8 cell layers in advance of the fungus hyphae. Hyphae were abundant on the epidermis, but never penetrated the host without first discoloring the host cells. Staling products were filtered from cultures of *Rhizoctonia*. Soybean seeds were germinated in the extracts, or the stems of aseptically grown soybean seedlings were wrapped with narrow strips of cotton soaked with the extract. Staling products alone, the diffusible substances produced by the fungus, reduced germination of soybean seed, inhibited development of secondary roots, caused necrotic lesions on stems and roots of seedlings, and killed seedlings within 7 to 10 days. Necrotic lesions produced by the extracts resembled lesions caused by hyphae of *Rhizoctonia*, but *Rhizoctonia* could not be isolated from them. No necrotic lesions appeared on control plants. (Minnesota Agricultural Experiment Station and United States Department of Agriculture.)

A three-spray schedule for the control of black rot of grapes. BRAUN, ALVIN J. Tests conducted in New York State vineyards indicate that under most conditions, black rot, *Guignardia bidwellii* (Ell.) Viala and Ravaz, can be effectively controlled by 3 applications, immediately before bloom, immediately after bloom, and 10–14 days later. Under conditions of severe disease occurrence, the percentage of control obtained with the various materials tested was significantly less, indicating the necessity of an additional pre-bloom application in vineyards where the disease is likely to be severe. Two late-season applications did not increase the control in those plots that already had

received 3 applications. Fermate (ferric dimethyl dithiocarbamate) 2–100 was more effective than any of the other fungicides tested, including Bordeaux 8–8–100. Bordeaux mixture caused injury to the foliage characterized by premature yellowing, especially in those plots receiving 5 applications of the 8–8–100 concentration. The foliage in the plots sprayed with Fermate retained a dark green healthy appearance, even after the foliage of the check plots showed signs of advanced maturity.

Correlated inheritance of resistance to eight races of wheat leaf rust, Puccinia rubigo-vera tritici, powdery mildew, Erysiphe graminis tritici, and glume color. CALDWELL, R. M., and L. E. COMPTON. The inheritance of seedling resistance to leaf rust races 9, 31, 65, 78, 79, 80, 101, and 110 and to two cultures of powdery mildew was studied by means of greenhouse inoculations, in F_3 and bulk F_4 progenies of individual F_2 plants of the cross Wabash C.I. 11384 × Michigan Amber 29–1–1–1, C.I. 4770. Rust reactions were determined on the first and mildew reactions on the third and fourth leaves of seedlings. The first leaf of Wabash gives either the resistant or "X" reaction to each of the 8 races. The "X" reaction usually changes to a resistant type in the second or third leaves. Michigan Amber was resistant to the two cultures of powdery mildew studied. Wabash has white glumes and Michigan Amber, brown. The segregation indicated a monogenic inheritance of resistance to leaf rust. Susceptibility was dominant. Each progeny reacted uniformly to the group of 8 races indicating that for all 8 races the same gene controlled either the resistant or "X" reaction. Mildew resistance and brown chaff color appeared to be monogenic and dominant. Leaf rust and mildew resistance and chaff color were independently inherited, the deviation from a phenotypic ratio for 3 factors being insignificant. (Purdue Agricultural Experiment Station and U. S. Department of Agriculture.)

Inheritance of resistance to loose smut of wheat, Ustilago tritici, in the varietal cross Trumbull × Wabash. CALDWELL, R. M., and L. E. COMPTON. During the 1939 and 1940 field seasons, respectively, 101 and 98 F_2 plants of the cross, Wabash × Trumbull, were inoculated by the partial-vacuum technique with a culture of loose smut to which Trumbull is resistant and Wabash susceptible. The F_3 progenies of the inoculated plants were grown in the field the years following inoculation when the percentages of heads infected were recorded. In both years approximately 75 per cent of the F_3 families fell in the range of 0 to 5 infection with the remainder distributed over the approximate range 5 to 85 per cent infected heads. F_2 populations of the same cross grown from seed from inoculated F_1 plants have produced only occasionally infected plants. Reciprocally crossed seed of the parentage, Wabash × Trumbull, containing the F_1 embryos have produced smut-free F_1 plants with few exceptions when inoculated by hand with a smut suspension. Backcrossed seed of the parentage Wabash ♀ × (Wabash × Trumbull) ♂ and inoculated with smut suspensions have produced partially infected progenies. These results suggest that Trumbull's resistance is dominant and monogenically inherited and that infection of the susceptible progeny of heterozygous resistant plants is prevented by the covering parental tissues of the ovary integuments or nucellus. (Purdue Agricultural Experiment Station and U. S. Department of Agriculture.)

Studies on the nature of "segregation" in certain plus strains of Glomerella. CHILTON, S. J. P., and H. E. WHEELER. Ascospores isolated from certain perithecial and nonconidial strains of *Glomerella* known as plus produced a high percentage of new strains known as minus. Occasionally, conidial but nonperithecial strains were found in the ascospore progenies. Analyses of asci gave ratios of 8 plus, 4 plus and 4 minus, or 8 minus cultures. The percentages were 8, 60, and 32, respectively. Only one ascus ratio occurred in a perithecium. Appropriate crosses among these strains indicated the new strains to be genetically different from the plus parental strain, the minus differing at one locus and the conidial at another. The two loci were apparently loosely linked. Ascospores of the plus strain were uninucleate. The evidence indicates that the new strains arose by mutation from the plus strain and the ascus ratios were determined by the nuclei entering and pairing in the ascogenous hyphae and fusing in the ascus.

Transfer of wildfire resistance from Nicotiana longiflora to N. tabacum. CLAYTON, E. E. A selection of *Nicotiana longiflora* was immune from the wildfire (*Phytomonas tabaci*) disease, while cultivated tobacco was susceptible. After prolonged efforts a cross was successfully made in 1940. Tetraploidy was not involved and progeny of the first backcross to tobacco (N.t. × N.l. × N.t.) were self- and cross-fertile. Inoculation and selection work produced an F_6 in 1944 that appeared to possess *N. longiflora* immunity from wildfire. This genotype was designated as T.I. 106. Tests were conducted with T.I. 106 in 1945. Inoculations with *P. tabaci* were begun with seedlings when they appeared above ground in the plant bed and continued until the end of the growing season in the field. No infection was secured. The tests were repeated in 1946 with the same

results. Adjacent checks of common tobacco were 100 per cent severely affected. T.I. 106 was crossed with varieties of commercial tobacco and 5 F_1's were planted in 1946. The F_1's reacted the same as the T.I. 106 parent, hence resistance was dominant. All F_1's showed marked heterosis. Increases in green weight ranged from 25 to 51 per cent, the comparisons being made with the commercial tobacco parents, which were larger than T.I. 106. Inoculation tests with *P. angulata* indicated that T.I. 106 was resistant to blackfire as well as wildfire.

The "dodder graft," a new method of using dodder to transmit plant viruses. COCHRAN, G. W. The "dodder graft" was devised to improve the dodder method of transmitting viruses. Stocks were rooted healthy plants, while scions were detached diseased stems or leaves. Stems of stock and scion were usually placed together and connected by detached dodder tips wound in a circular or figure-eight manner. Winding was facilitated by using a paste of Carbowax 4000 with 15 per cent water. An inch or more of the stems were wound and tied with string. The Carbowax was then removed with water. The "dodder grafts" were sprayed with a mixture of Fermate, Omilite, and AO_3 to prevent a rotting caused by an unidentified *Botrytis sp.* Then they were continuously atomized with water under incandescent lights for 10 to 14 days. The dodder usually made haustorial connections to both stock and scion in 3 to 4 days. The stocks were shaded on the fifth day and sprayed with 100 p.p.m. of indole butyric acid on the seventh day. The scion and connecting dodder were removed on the tenth to fourteenth day. When the "dodder graft" method is used, virus transmission is improved because of the increased number of shortened dodder stems connecting the diseased and healthy tissues.

Injury to root tissue caused by plant residues. COCHRANE, VINCENT W. The decomposition products of plant residues have been described as directly or indirectly injurious to plant roots. Direct injury to root tissue can be assayed by exposing the radicle of 72-hour-old radish seedlings to the action of cold-water extracts of the ground plant tissue. Extracts made from undecomposed ladino clover and from rye grass caused browning of radish roots in 8 hours. Decomposition of these residues at 2 moisture levels resulted in a progressive disappearance of the factor causing the injury, although microbial action brought about temporary increases in toxicity. Decomposition of ladino clover tissue at 56° C. accelerated destruction of the toxic factor, presumably as a result of the activity of thermophilic bacteria. Apart from this case, studies of microbial populations in the decomposing residues failed to establish any significant association of particular groups of microorganisms with the appearance or disappearance of root injury. Extracts from nondecomposed corn stover did not cause root browning; microbial action under anaerobic conditions caused a transitory appearance of injurious activity. Extracts of soybean tissue did not cause discoloration of radish roots, whether tested before, during, or after decomposition.

A method of forecasting late-blight epiphytotics in Eastern Virginia. COOK, HAROLD T. Late-blight epiphytotics have occurred on early potatoes in Eastern Virginia in only 2 of the last 17 years (1930–46) and on tomatoes in but one year. Routine use of fungicides has not been justified because of the infrequent occurrence of blight, but spraying or dusting would be valuable in blight years if epiphytotics could be predicted. Analysis of meteorological data for the 17 years show that epiphytotics may be predicted by plotting the average weekly temperature and cumulative rainfall for May and June on ordinary cross-section paper, and that the advisability of spraying may be determined by this means at weekly intervals during the growing-season. These data show that the development of epiphytotics is dependent upon a combination of above normal rainfall and temperatures below 75° F. from the middle of May to at least the middle of June. Such a combination occurred only in 1938 and 1946, when blight was prevalent. Either rainfall or temperature, or both, were unfavorable for blight in the other 15 years. The tomato crop was damaged only in 1946 when temperatures below 75° F. continued through June.

Occurrence of Actinomyces antibiotic to Pythium in some sugar-cane soils of Louisiana. COOPER, V. E., and S. J. P. CHILTON. In studies initiated in the sugar-cane area of Louisiana on the Pythium root rot of sugar cane, a survey was made in 1946 of the antibiotic *Actinomyces* present in the soils of the cane area. Of 3788 cultures tested against a parasitic strain of *Pythium* and classified according to the distance at which the *Pythium* was inhibited in Petri-dish tests, 896, or 23.6 per cent, showed some antibiosis. Of these 365 inhibited *Pythiums* at 1 to 5 mm.; 301 inhibited at 6 to 10 mm.; 140 at 11 to 15 mm.; 90 at 16 or more mm.; and only 10 inhibited at 35 or more mm. A progressive increase of the Actinomyces populations occurred from March to August, but the percentage of antibiotic strains in the samples tested decreased. The percentage

of antibiotic strains and the average antibiosis of the strains was lower in heavy or root-rot soils than in the light soils.

A bacterial storage disease of sweet potatoes. COX, C. E., and D. W. SQUIRES. An apparently undescribed disease of Maryland Golden sweet potatoes was observed in an experimental storage house on the University of Maryland farm in 1945. A possibly identical storage rot has been reported from a commercial storage house elsewhere in the State. The disease typically involved the entire root, which, externally, became chocolate-brown in contrast with the orange color of healthy sweet potatoes. Diseased potatoes were flaccid and flexible but not mushy. Internally, they were grayish-brown and water-soaked, but the cells did not separate as in soft rot. The odor was distinctive and acrid rather than putrescent. Unless attacked by secondary invaders, the affected roots eventually dried out and shriveled to form hard mummies. A bacterium has been isolated from diseased tissues. This organism is capable of reproducing symptoms of the disease in sweet-potato slices inoculated in the laboratory. It did not cause decay of slices of white potato, tomato, onion, or carrot inoculated similarly. The organism is a short rod, truncate at the ends, nonmotile and Gram-negative. It grows slowly and sparsely on potato-dextrose and malt-extract agars, but makes rapid and fairly abundant growth on sweet-potato-decoction agar.

Specificity of certain organic nitrogen compounds for bacterial plant pathogens. DAVIDSON, RICHARD S. The bactericidal activity of four chemical groups comprising 15 quaternary nitrogen compounds was determined for the following bacteria associated with plant diseases: *Aerobacillus polymyxa, Bacillus mesentericus, Erwinia carotovora, Phytomonas michiganensis, Corynebacterium sepedonicum,* and *Bacterium solanacearum.* The groups, in the order in which they produced 100 per cent kill *in vitro* of 24-hour broth cultures, after 10 minutes exposure, were quinoliniums, pyridiniums, nicotiniums, and quaternary ammoniums. Lauryl isoquinolinium bromide was the most active of the group as a bactericide. The least potent of the group was di-methyl di-lauryl ammonium thiocyanate. *A. polymyxa* and *B. mesentericus,* both spore-formers, were killed by higher dilutions of the compounds included in the test than were the nonspore-formers. Of the nonspore-formers included in this study, the Gram-positive bacteria, *P. michiganensis* and *C. sepedonicum,* were killed by higher dilutions of the compounds studied than were the Gram-negative bacteria, *E. carotovora* and *B. solanacearum.*

Control of late blight of tomato in New Jersey. DAVIS, B. H., and C. M. HAENSELER. Late blight, caused by *Phytophthora infestans,* became prevalent in 1946 on tomato foliage in canhouse areas the first week in July. Almost continuous spread followed, the peak of fruit destruction occurring August 20 to 25. Spraying experiments were started in a commercial field 3 days after scattered foliage infection was noted. Eight applications were made at 10-day intervals using a 5-row power take-off sprayer with 3 nozzles per row. Microgel, a tribasic copper sulphate containing 50 per cent copper (4–100), Fermate (ferric dimethyldithiocarbamate) (2–100) alternating with Microgel (4–100), and Zerlate (zinc dimethyldithiocarbamate) (2–100) each containing calcium arsenate (4–100) were used. Control plots received calcium arsenate and lime (4–8–100). Yield and disease data were taken on 100 plants in each treatment. At weekly pickings all ripe cull fruits and rotted green fruits were picked, counted, and discarded. All remaining ripe fruits were then picked, weighed, counted, and marketed. Calculated yields in tons of marketed fruits per acre were: Microgel, 27.7; alternating sprays, 21.4; Zerlate, 13.8; control, 10.2. Of these marketed fruits, 0.2, 2.0, 13.3, and 10.8 per cent were slightly infected. Of the total fruits produced 0.7, 12.1, 55.9, and 59.8 per cent were infected.

Some factors influencing inoculation of tobacco with streak virus. DIACHUN, STEPHEN. Leaves slightly below the primary growing point of systemically infected plants of *Nicotiana rustica* were a more reliable source of tobacco-streak virus than tobacco or sweetclover tissue. Local lesions developed on every one of several hundred tobacco leaves dusted with 600-mesh carborundum and rubbed with a glass spatula dipped in *N. rustica* tissue crushed with M/10 Na_2HPO_4. Without the M/10 Na_2HPO_4 and carborundum the number of local lesions was greatly reduced. Dilution of the inoculum with M/10 Na_2HPO_4 beyond 1: 2 decreased the number of local lesions, but the inoculum was still effective at a dilution of 1: 32. Additional rubbing, up to a certain point, increased the number of local lesions. Leaves supported on a flat surface during rubbing did not develop more lesions than leaves merely held at the tip. Very young and old leaves were not so susceptible as vigorous medium-young leaves (5th or 6th below growing point), especially on relatively small plants.

Reaction of 35 species of Nicotiana to tobacco-streak virus. DIACHUN, STEPHEN, and W. D. VALLEAU. None of the approximately 200 accessions of tobacco tested seemed sufficiently resistant to the tobacco-streak virus to be useful as breeding stock. Thirty-five of the approximately 60 described species of *Nicotiana* have been inoculated. The reaction ranged from a high degree of resistance to extreme susceptibility. Most of the species were moderately susceptible, similar to *N. tabacum.* In *N. bethamiana, N. clevelandii, N. mcgalosiphon,* and *N. miersii* systemic infection caused severe necrosis, often resulting in death. *N. glauca* and *N. gossii* seemed sufficiently resistant to have value as breeding material.

Symptoms of Dutch elm disease reproduced by toxins of Graphium ulmi in culture. DIMOND, ALBERT E. *Graphium ulmi,* grown on synthetic media, either in shake or quiet submerged culture, produces at least 2 metabolites toxic to elm and tomato cuttings. Each toxin produces symptoms typical of a part of the syndrome of Dutch elm disease. These toxins, produced in 1 month in quiet culture, are as abundantly produced in 4 days in shake culture. The raw culture is filtered through paper, evaporated *in vacuo,* and centrifuged to remove mycelial fragments. The highly toxic clear supernatant liquid in 70 per cent alcohol precipitates a gummy amorphous compound. This is a polysaccharide which gives the red color of a dextrin with IKI, and is hydrolyzed to reducing sugar in hot 1 per cent H_2SO_4. On tomato and elm cuttings it produces an upcurling of leaflets and leaves with marginal withering of the blade, but causes no necrosis. The second fraction is soluble in 70 per cent alcohol, producing a striking interveinal necrosis on tomato and elm. Ether extraction of this fraction to remove organic acids does not alter its toxicity. The ether-insoluble, alcohol- and water-soluble fraction produces severe interveinal necrosis on tomato cuttings.

Culture techniques for large numbers of diseased and dead wood specimens. FATE, LESTON R., CLYDE E. DIKE, and O. N. LIMING. Standard techniques have been modified and new ones developed for handling several hundred specimens daily in the Dutch Elm Disease Identification Laboratory. Specimens from diseased but living parts of elm trees infected with *Ceratostomella ulmi* or other vascular parasites are cultured on a simplified potato-sucrose agar medium. The cultures are held for 3 days at 50° F. and then for 3 days at room temperature. Dead wood and bark materials are cultured by a wet-plate technique, employing excess water and low temperature incubation for 21 days and then room temperature for three days. Low temperature suppresses the growth of most other organisms, while *C. ulmi* is developing. The warm-up period, during which the coremia mature, is an important step in the technique. Through the use of antiseptic solutions, the upward direction of air currents, and low-temperature incubation, laboratory contaminations are satisfactorily controlled.

Identification of Ceratostomella ulmi in pure cultures and in mixed associations of organisms. FENNER, LAWRENCE M., and O. N. LIMING. Zonate colony growth and conidiophores with young spores form the basis for identification of *Ceratostomella ulmi* on potato-sucrose agar. Coremia may form on the wood plantings, but usually not on the agar medium. *Verticillium* and *Cephalosporium* isolations form different type colonies. In wet plate cultures, *C. ulmi* coremia have translucent white heads which may droop or sink over the stalk, and develop surface striations when exposed to dry air. Six other *Graphium* species have different stalk and head characteristics. Several saprophytes and air contaminants may over-run and mask *C. ulmi* mycelium and coremia. *Dothiorella ulmi* and a *Cytospora* limit *C. ulmi,* whereas *Sphaeropsis ulmi* and a *Dematium* favor *C. ulmi* coremial formation.

Pathogenicity of isolates of the onion pink-root organism. GORENZ, A. M. Various isolates of *Phoma terrestris* were tested in sand culture on Yellow Globe, a very susceptible variety, and Yellow Bermuda, a moderately resistant one. Severe damping-off along with the typical discoloration of the roots occurred on Yellow Globe seedlings with the 2 most virulent isolates, 1 from Texas and 1 from Louisiana. On Yellow Bermuda, damping-off was less common, but typical discoloration of the roots was present. Isolates from Utah, Colorado, Iowa, and Wisconsin were less virulent and caused only typical root discoloration. Pycnidial production on corn-meal agar was observed in the Louisiana, Texas, Iowa, and Colorado isolates. In plants infected with the Louisiana isolate mature pycnidia were produced abundantly on the roots, this being the first record of the development of pycnidia on the host. (University of Wisconsin and U. S. Department of Agriculture.)

Influence of methocel sticker on the effectiveness of Arasan for onion-smut control. GORENZ, A. M., and J. C. WALKER. Onion-seed-treatment experiments for the control of smut (*Urocystis cepulae*) were conducted in southeastern Wisconsin in 1946. Arasan

(tetramethyl-thiuram-disulphide) was used with and without methocel sticker. Seed was sown at approximately 65 lb. per acre, the usual rate for production of onion sets. In one series of experimental plots an average of 49.8 per cent of the plants from seed with no treatment were diseased. When $\frac{1}{10}$ lb. of Arasan per pound of seed was applied without sticker the percentage of diseased plants was 3.8. When $\frac{1}{8}$ lb. was applied with sticker there was a significant increase in disease to 9.8 per cent. When the dosage was increased to $\frac{1}{4}$ lb. with sticker there was a significant decrease to 2.5 per cent. In another series of trials on a commercial scale there was a similar increase of disease when $\frac{1}{8}$ lb. was applied with sticker over $\frac{1}{10}$ lb. without sticker. In commercial practice results with $\frac{1}{10}$ lb. without sticker were variable, while with sticker more uniform results were secured but a dosage of $\frac{1}{4}$ lb. was necessary for maximum control. (University of Wisconsin and U. S. Department of Agriculture.)

Growth of Streptomyces griseus in shake-flasks. GOTTLIEB, DAVID, and H. W. ANDERSON. The growth of *Streptomyces griseus* was studied by 3 methods, (1) microscopic observation, (2) mycelium weight, and (3) viscosity measurement. Exact time of spore germination was difficult to determine, since, even under oil immersion, only an elongation of the spores was observed. At 6 hours the mycelium was sparse and consisted of some small individual hyphae and of longer branched hyphae which tended to round up into a loose weft. With increased growth, the weft formed masses of mycelium consisting of a dense solid center and a periphery of branched radiating hyphae. At 24 to 30 hours the entire body of medium was filled with these clumps. The peripheral hyphae from the central cores intermingled and produced a continuous mass of intertwining hyphae. The culture appeared very viscous at this stage. After 48 hours the mycelium began to fragment and spores were produced. The number of spores increased, and at 84 hours a lysis of the mycelium was apparent. The dense central core also disintegrated into granular pieces. The culture became less viscous during this stage. Both viscosity and mycelial weight determination showed an increase, to a maximum at 24–30 hours, then a decrease up to about 96 hours followed by a gradual levelling.

Investigations on grey speck of oats in Manitoba. HAGBORG, W. A. F. A disease, thought to be grey speck, was found in varietal plots at Winnipeg and in farmers' fields at several other localities in Manitoba. Pot experiments in 1944 furnished proof that the disease was grey speck and in subsequent field experiments statistically significant increases in yield were obtained following the use of manganese sulphate. When manganese sulphate was applied to the soil (65 lb. $MnSO_4$/acre) the increase in yield was 58 bu./acre; when applied as a spray (9 lb. $MnSO_4$/acre) it was 24 bu./acre; when applied as a 25 per cent dust diluted with clay (8 lb. $MnSO_4$/acre) it was 20 bu./acre; and when applied as a seed steep (9.5 lb. $MnSO_4$/acre) it was 12–17 bu./acre. Marked differences were observed in varietal susceptibility to grey speck. In a replicated test of 68 varieties in 1946, *Avena strigosa*, Black Mesdag, Ajax, Exeter, and Laurel were among the more resistant varieties, while Victoria, Tama, Vicland, Bonda, Early Miller, Valor, Sixty-Day, Erban, Trispernia, Mindo, Legacy, and Bond, were among the more susceptible. High susceptibility has been noted among varieties descended from Victoria, but there is not a complete linkage between resistance to stem and crown rusts and susceptibility to grey speck.

A plot inoculation method for determining the resistance of wheat varieties to bacterial black chaff. HAGBORG, W. A. F., and R. F. PETERSON. A simple method of inoculation for initiating infection with bacterial black chaff in field plots was found effective in a yield test of 98 varieties of wheat. After spraying the plots with an aqueous suspension of *Xanthomonas translucens* f. sp. *undulosa* (S.J. & R.) Hagb., a cloth, moistened with the suspension, was dragged over the plants. Three of the 6 replicates were inoculated and 3 left noninoculated. Comparable means for inoculated and noninoculated plots, respectively, were as follows: leaf infection, per cent, 39.7, 5.1; head infection, per cent, 18.7, 8.4; yield, bu./acre, 39.4, 43.3. The 3 differences were statistically significant ($P < 0.05$). Leaf and head infections were significantly correlated: varietal means $r = 0.52$ ($P < 0.01$), plots within varieties $r = 0.38$ ($P < 0.01$). Analyses of variance of the data from the three inoculated replicates, with the varieties considered in 10 groups based on ancestry, established the following significant differences between varietal means: leaf infection, all varieties $P < 0.01$; leaf infection, varieties within each of 9 groups $P < 0.05$; head infection, all varieties $P < 0.01$; head infection, varieties within each of 4 groups $P < 0.05$. Differences between group means were highly significant: leaf infection $P < 0.01$, head infection $P < 0.01$. A sound basis existed, therefore, for selecting varieties resistant to bacterial black chaff. (University of Manitoba and Dominion Laboratory of Cereal Breeding.)

A new disease of garden pea. HARE, WOODROW W. A species of *Pythium* was isolated from dead tips of garden pea (*Pisum sativum*) in 1941 and its pathogenicity to

young foliage of peas determined. This disease caused 5–10 per cent loss in experimental plantings in 1946, and isolations gave the same *Pythium*. In the field, infection occurs in the axils of young leaves or in the bud of the plant. A single leaf may be killed but usually the entire tip is killed by infection in the bud or from the leaf axil. Affected tissues collapse quickly and become dark-brown. Only the upper 3 to 5 internodes are killed in most cases and new branches are put out below. Greenhouse inoculations of potted plants with mycelial-fragment suspensions in buds or leaf axils gave similar symptoms. The disease has been seen only after periods of heavy rain.

Heptadecylglyoxalidine (341) and its hydrolysis product as fungicides. HARRY, JOHN B., R. W. MCNAMEE, and R. H. WELLMAN. In 1946, 2-heptadecylglyoxalidine was melted and slurried in hot water to provide a dispersible paste. In previous years it had been formulated as a hydrochloride mixed with pyrophyllite, or as a cold, wet, ball-milled product. The 1946 slurry failed to control apple scab and controlled cherry leaf spot less effectively than did previous products. It was found that under the conditions of preparation of the slurry, the glyoxalidine had hydrolyzed. The hydrolysis product is less effective than the parent glyoxalidine in laboratory spore-germination tests and greenhouse disease-control tests. It can be distinguished from the glyoxalidine by spectral analysis or by its biological effectiveness. The wet ball-milled slurry hydrolyzes much more slowly than the hot-mixed slurry produced in 1946. In the time lapse between wet ball-milling and field application in 1942, the glyoxalidine was only partly hydrolyzed, and good control of apple scab was obtained. The hydrochloride was not hydrolyzed at all. Past results have been best when the hydrochloride was used. (Crop Protection Institute and Carbide and Carbon Chemicals Corporation.)

Zinc ethylene bisdithiocarbamate as a fungicide on vegetables. HEUBERGER, J. W., S. H. DAVIS, JR., L. P. NICHOLS, and L. D. BUEHLER. In 1946, zinc ethylene bisdithiocarbamate (reaction product, coded He 178e) was tested as a spray at the rate of 1½–100 active ingredient on potatoes, tomatoes, cucumbers, cantaloupes, and celery to evaluate its disease-controlling powers, phytotoxicity, and effect on yield. *Disease control:* Of all the copper and organic fungicides tested (Bordeaux, Compound A, Tribasic, Yellow Cuprocide, Zerlate, Dithane + zinc sulphate-lime, CPI 169A, CPI 341) zinc ethylene bisdithiocarbamate gave the best control of early and late blight diseases on potato and tomato, of anthracnose fruit spot on tomato, of cucumber downy mildew, and of Cercospora leaf spot on celery. On cantaloupe it was slightly inferior to Yellow Cuprocide for downy mildew control and to all copper compounds for powdery-mildew control. *Phytotoxicity:* Zinc ethylene bisdithiocarbamate was nonphytotoxic to potato, tomato, and celery. It improved color on cantaloupe and cucumber but caused slight reduction in leaf size. *Yield:* Zinc ethylene bisdithiocarbamate gave the highest yield of potatoes, tomatoes, cucumbers, and cantaloupes; celery yields were not obtained. A second formulation of zinc ethylene bisdithiocarbamate (coded IN 5446) was also tested at equivalent active ingredient concentration to He 178e. It was as effective in disease control and yield response as He 178e, but was somewhat more toxic on cucumber and cantaloupe.

New organic fungicides and insecticides for potatoes. HEUBERGER, J. W., and L. A. STEARNS. Research in 1945 and 1946 on use of copper and new organic fungicides (Bordeaux, Compound A, Zerlate, Dithane plus zinc sulphate-lime, zinc ethylene bisdithiocarbamate) and insecticides (DDT, Gammexane, Rhothane, X3956) for disease and insect control showed that: (1) DDT was the outstanding insecticide for potato leafhopper control; (2) DDT was noninjurious to potatoes; (3) DDT gave high yield responses when leafhoppers were present but not when they were absent; (4) DDT has little or no fungicidal value; (5) DDT and the copper and organic fungicides were compatible since each material was as effective when used in combination as when used alone; (6) Zerlate (zinc dimethyl dithiocarbamate) gave excellent control of early blight but failed to control late blight satisfactorily; (7) Dithane (disodium ethylene bisdithiocarbamate) plus zinc sulphate-lime gave excellent control of both early and late blights and high yield response; (8) Zinc ethylene bisdithiocarbamate (reaction product-He 178e; IN 5446) gave the best control of both early blight and late blight, and the highest yield of potatoes, of any fungicide used; (9) a combination of zinc ethylene bisdithiocarbamate and DDT was the most effective combination of any used for joint disease and insect control and gave the highest yield response.

The toxicity to tomato cuttings of several microbial and other polysaccharides. HODGSON, ROLAND, W. H. PETERSON, and A. J. RIKER. The work on wilting induced by .. glucosan from crown-gall bacteria has been extended to include several other polysaccharides. All these (in 0.4 per cent solution) produced in tomato cuttings a wilting, and, in addition, some caused irregular necrotic areas in the leaflets. Symptoms were of two general types: (1) a wilting and necrosis of the leaflets, and (2) a wilting primarily

of the stems. Intermediate effects were sometimes encountered. Toxicity of the first type appeared with the following: glucosan from crown-gall bacteria, inulin, soluble starch, fructosans (*Azotobacter indicum* and *Bacillus subtilis*), dextran (*Bacterium* sp.), a polysaccharide from black spruce, and three water-soluble, corn-syrup dextrins of different molecular size. Symptoms of the second type were shown by a gum preparation (*Rhizobium trifolii*), dextrans (*Leuconostoc mesenteroides, Leuconostoc dextranicum,* and an unidentified soil organism), and an amylo-dextrin preparation. Factors appearing important in the type of symptoms induced are solubility and molecular size of the polysaccharide. The water-soluble polysaccharides of low molecular weight induced wilting of the first type. Since all of these preparations were toxic, it appears that other polysaccharides may induce similar symptoms, and that wilt-inducing plant pathogens, particularly, should be examined for substances that act in a similar way.

Parasitism of Actinomyces scabies on various plants. HOOKER, W. J. Seedling plants representing 8 families developed root necrosis when grown in soil-extract agar artificially infested with pure cultures of *Actinomyces scabies.* A group of 10 cultures of *Actinomyces sp.* were tested on seedlings of wheat, garden pea, soybean, corn, radish, and cucumber, and on potato sprouts. Six of the 10 cultures caused neither appreciable necrosis of potato stems nor injury to seedling roots. The remaining 4 cultures, 3 of which scabbed potato tubers severely, caused necrosis of potato stems in a manner typical of *A. scabies.* These same four cultures caused severe necrosis of roots as well as a reduction in root weight with wheat, pea, soybean, and radish. Necrosis was most pronounced on root tips, and the development of secondary roots was almost inhibited. Corn roots were only slightly necrotic but their weight was markedly reduced, the most noticeable reaction being a slight thickening of the secondary roots. Roots of seedling plants of barley, oats, onion, tomato, eggplant, squash, red beet, carrot, parsnip, and Lima bean became necrotic when tested with one pathogenic culture of *A. scabies.* Cucumber roots were apparently unaffected by any of the 10 cultures.

Stem necrosis of potatoes caused by Actinomyces scabies. HOOKER, W. J., and G. C. KENT. Additional information is presented concerning parasitism of *Actinomyces scabies* on potato stems. Brown, necrotic lesions on subterranean Cobbler stems originating at lenticels or at points of emergence of stolons and secondary roots were obtained in greenhouse experiments using artificially infested peat soil. In advanced cases, the stem was girdled and rotted at the base with vascular discoloration extending up the stem 6 to 8 internodes. Terminal leaflets were rolled upward, chlorotic, purple to red, coloration beginning at base and progressing toward tip. Lesions caused by *A. scabies* were lighter brown than those typically caused by *Rhizoctonia solani,* with the margin not so clearly defined. In such necrotic tissue the organism consistently was demonstrated histologically and successfully isolated. In a test of 10 potato progenies, varieties resistant to tuber scab were likewise resistant to stem necrosis. In the summers of 1945 and 1946 in peat soils of northern Iowa well-developed, somewhat circular lesions were present on stems of resistant and susceptible potatoes with *A. scabies* sporulating on the surface and with its filaments well established in the tissues.

Two new fungicidal molecular configurations. HORSFALL, JAMES G., and RAYMOND W. BARRATT. The fungicidal action of 4-nitrophenyl-2,3-dichloroisobutyl ether, $4\text{–}NO_2C_6H_4OCH_2CCl(CH_3)CH_2Cl$ (code Cr 1520) was discovered in the laboratory in 1945, and the action of 1 hydroxy-2-trichloroethyl bis-2-chloroethyl phosphite, $(ClCH_2CH_2O)_2POCH(OH)CCl_3$ (code Cr 1432), in 1946. Both were compared in the field as sprays at 2½ lb. per 100 gal. with disodium ethylene bisdithiocarbamate (code D) and 2,3-dichloronaphthoquinone (code P). Percentage control = 100 – (disease in check × 100/disease in treated) for the two doses was as follows: *Cercospora apii* in 1945—Cr 1520—68 and 32, D (1 lb. only)—97.5; in 1946—Cr 1432—42 and 39, D—92 and 88, P—94 and 92; *Septoria apii* in 1946—Cr 1432—35 and 35, D—94 and 91, P—98 and 92; *C. lindemuthianum* in 1946—Cr 1520—62 and 57, D—76 and 62, P—86 and 67; also in 1946—Cr 1432—73 and 52, D—83 and 62, and P—80 and 42. Whether improvement in conditioning or in molecular structure will lift the performance of these new materials remains to be seen. Differences for Cr 1432 between beans and celery already suggest specificity. Thanks are due Rohm and Haas Co. for the chemicals.

An organic cadmium fungicide for turf diseases. HOWARD, F. L., and H. L. KEIL. Para-aminophenyl cadmium dilactate has given superior control of the four principal diseases of Creeping and Velvet Bent turf, namely: dollar spot (*Sclerotinia homoeocarpa*), copper spot (*Gloeocercospora sorghi*), brown patch (*Corticium vagum*), and pink patch (*Corticium fuciforme*). Comparison with 20 fungicidal chemicals in randomized replicated plots for 2 seasons has shown the cadmium complex to be nonphytotoxic, and to have unusual disease-arresting and residual fungitoxicity. Dosages of 0.1 lb. of wettable

powder containing 20 per cent toxicant in 10 gal. water (1: 4000) applied to 1000 sq. ft. have proved effective. Two applications retarded copper spot development on Piper Velvet Bent to a trace, whereas control areas developed 24 per cent disease. An application to Narragansett Creeping Bent with 7 per cent area killed by dollar spot permitted filling in by new growth within 10 days in contrast to increased disease in the nontreated area.

Vulcanized saponifiable oils (factices) as fungicide deposit builders. HOWARD, FRANK L., and HUGH H. MOSHER. A common weakness of fungicides is lack of tenacity on plant surfaces and inability to build up a sufficient deposit of the toxicant to compensate for this. Fat-sulphur condensates made by reacting saponifiable fats and oils with sulphur have been found which will permit spray residues of water-soluble, surface-active toxicants to adhere to plant surfaces despite washing. Vulcanized rapeseed, soya, corn, castor, linseed, and other vegetable oils are nonphytotoxic at effective concentrations and decrease the injuriousness of some fungicides. While these factices can be added separately to the pesticidal spray, at the rate of 0.1 lb. to 1.0 lb. per 100 gal. water, a homogenous, stable, combined solution can be obtained. DDT and similar oil-soluble insecticides can be added to the formulation. A preparation of rapeseed factice with lauryl isoquinolinium bromide as the toxicant has given an L.D. 98 at 1 p.p.m. against *Macrosporium sarcinaeforme*. The residue from a water drop containing 1: 5000 of the toxicant can be washed more than 30 minutes and still inhibit germination better than 85 per cent of *M. sarcinaeforme* spores. (Rhode Island Agricultural Experiment Station and Onyx Chemical Co.)

The effect of some of the newer fungicides on yield of potatoes in the absence of early and late blight. HOYMAN, WM. G. Using a triple lattice design with 6 replications, 6 applications of certain fungicides were applied at 10- to 12-day intervals from July 12 to September 3, 1946, to Bliss Triumph potatoes at Grafton, North Dakota. The manufacturers' recommendations were followed with respect to the concentrations of the materials. In order to reduce the damage from insects, 1 lb. of actual DDT was added to each 100 gal. spray material and the materials applied at the rate of 150 gal. per acre at 400 lb. pressure. Dusts were applied at the rate of 40 lbs. per acre and included 5 per cent of actual DDT. In the absence of early and late blights, the dust treatments and their respective yields in bushels per acre were as follows: He 178, 165; Copper A, 147; Zerlate (zinc dimethyldithiocarbamate), 169; Tribasic Copper Sulphate, 156; DDT, 162 and DDT, 166. The various materials used as sprays with their respective yields were: Phygon (2,3-dichlor-l,4-naphthoquinone), 164; Tribasic Copper Sulphate, 162; Bordeaux Mixture, 172; Dithane D 14, 168; He 178, 154; Polyethylene Polysulphide (Omilite), 133; Polyethylene Polysulphide plus Phygon, 177; Zinc Ethylene Bisdithiocarbamate, 183; and Manganese Ethylene Bisdithiocarbamate, 153. The plots receiving no treatment yielded 138 bushels per acre.

Structures corresponding to appressoria and substomatal vesicles produced on nutrient-solution agar by germinating urediospores of cereal rusts. HURD-KARRER, ANNIE M., and H. A. RODENHISER. Germ-tubes from urediospores of *Puccinia graminis tritici*, *P. triticina*, *P. dispersa*, *P. coronata avenae*, *P. hordei* (*simplex*), and *P. sorghi* on the surface of agar made with a mineral nutrient solution and glucose produced bodies whose mode of development, size, and shape identify them as structures corresponding to appressoria and substomatal vesicles. They had, for the most part, the size, distinctive shape, septation, and number of infection hyphae that characterized the substomatal vesicle of the particular species as described by Pole-Evans from stained sections of their respective host plants. Infection hyphae were never observed to develop beyond a length of about 300 μ.

Graft-transmissible brooming disease of walnut. HUTCHINS, LEE M., and HORACE V. WESTER. Brooming of black walnut (*Juglans nigra*), butternut (*Juglans cinerea*), and Japanese walnut (*Juglans cordiformis* var. *ailantifolia*) has been observed over several years in parts of the eastern United States. It is characterized mainly by brooms or sucker growth on main stems and branches, tufting of terminals, profusion of branchlets from axillary buds, dwarfing of leaves, and sometimes by death of the trees. Symptoms vary from mild to severe on different trees of these species and are particularly pronounced on Japanese walnut. Inoculation experiments employing patch-bark grafts, performed in 1944 and 1945, demonstrated transmissibility of the brooming disease in black walnut and in Japanese walnut, the incubation period varying from several months to two years, under conditions of the experiments. Cross inoculations among *Juglans* species are not sufficiently mature for a report at this time. Transmission of the brooming disease by grafting and absence of a visible pathogen indicate probable virus causation. (U. S. Department of Agriculture and U. S. Department of the Interior.)

Virus attenuation and mutation. JOHNSON, JAMES. Sea-holly (*Eryngium aquaticum*) inoculated with ordinary tobacco mosaic virus, which yields severe symptoms on tobacco, results in only mild symptoms on tobacco after passage through sea-holly. Single-lesion strains of the severe and mild forms of the disease may be isolated and pure-lined on *Nicotiana glutinosa* or the *glutinosa-tabacum* hybrid, by repeated dilution. The severe strain usually produces a large spreading lesion and the mild form a smaller and more localized lesion on the latter hosts. The severe strain is, however, more localized and slower moving in the sea-holly than is the mild strain. Thus this host acts as a "filter" host separating the two-related strains in question, and yields what is commonly referred to as an attenuated strain without any true mutation occurring in the sea-holly. Single-lesion severe strains have, however, been changed so as to yield mild forms by growing the virus on tobacco at a temperature of 36–37° C. for several days. The latter is regarded as mutation to a mild form which can be isolated either by the local lesion method or by passage through a host such as sea-holly, yielding a pure attenuated virus. Thus mutation of the tobacco mosaic virus normally precedes attenuation. The terms "attenuation" and "mutation" are frequently used loosely in the literature; whereas they be quite definitely applied in the results with *Eryngium*, when tested by the pure-lined single-lesion method.

Fungus invasion of water-congested tissues. JOHNSON, JAMES. Several plant species were grown in the greenhouse on sandy soils low in available potash. Part of each series was transferred outdoors for 5–10 days. The indoor and outdoor plants were then inoculated with typical fungus parasites by atomizing without wounding. They were placed in a moist chamber with controlled soil and air temperatures favoring water-congestion in the leaves. The moist-chamber exposures usually varied between 12 and 24 hours. The outdoor plants commonly water-congested first and most extensively and were usually more rapidly and heavily infected by the fungus parasites than greenhouse-grown plants. Frequently, susceptible varieties water-congested more easily than resistant varieties grown in the same container. When resistant varieties became congested, they often developed heavy infection. The results varied with the time required for congestion and allowed for spore germination and infection. All factors concerned could not be controlled at will, and some uncertain results were obtained. The best correlations were secured with bean anthracnose, tomato late blight, leaf rust of oats, and leaf rust of wheat. The results with potato late blight and sunflower rust were more difficult to interpret because of failure to secure good visible signs of water-congestion.

The effect of hydrogen-ion concentration on the toxicity of Spergon. KELMAN, ARTHUR. A relationship between hydrogen-ion concentration and fungitoxicity of Spergon (98 per cent tetra-chloro-p-benzoquinone or chloranil) was demonstrated in the laboratory. Mycelial development of *Glomerella gossypii* and *Rhizoctonia solani* in nutrient solutions containing varying amounts of Spergon was suppressed at pH 3.8–4.8, whereas growth developed at pH 6, 7, and 8, reaching a maximum at pH 7. In order to obviate shifts in pH, discs of mycelium were exposed to suspensions of Spergon in buffer solutions adjusted to 6 pH levels. When the discs were removed to potato-dextrose-agar plates, slowest mycelial development occurred from discs taken from the most acid buffer solutions. In these tests the action was fungistatic rather than fungicidal. A distinct pink coloration usually developed above pH 4.8 in solutions containing Spergon, increasing in intensity as the pH level increased. In greenhouse experiments to determine if the phytotoxicity of Spergon in soil was influenced by hydrogen-ion concentration, the toxic effect on cotton seedlings was greatest in the most acid soil. The decrease in effective toxicity of Spergon with decrease in hydrogen-ion concentration is probably associated with the conversion of chloranil to a less toxic compound. Although variation in effective toxicity may occur in a normal pH range, the low solubility of chloranil precludes complete conversion or complete loss of toxicity.

Overwintering and infection of the tomato anthracnose organism. KENDRICK, J. B., JR. From a tomato field at Madison, Wisconsin, heavily infected with *Colletotrichum phomoides*, dead stems and petioles collected in late fall and early the next spring had inconspicuous black stromatic bodies that readily yielded the causal organism, showing that the latter overwinters in tomato refuse. Plants grown on the same field the following season were inoculated with a nonpigmented strain, isolated in Wisconsin in 1942 by W. J. Hooker, which served as a marker. Random isolation from affected fruits showed that practically all infection came from overwintering refuse and not from current season artificial inoculation. Greenhouse studies showed that the organism can infect the foliage of young tomatoes, peppers, and eggplants in a very humid atmosphere. At 28° C. lesions developed on tomato seedlings after 3 days of constant moisture, while 11 days were required at 16°. Further development of infection after the abscission of the infected cotyledons and leaves did not occur even at continued high moisture.

Current season infection of tomato foliage appears to be not so important in causing fruit infection as does the primary inoculum from tomato refuse of previous seasons.

Quick decline of orange trees. KLOTZ, L. J., and G. A. ZENTMYER. Quick decline has, with certainty, been found attacking only sweet orange trees (2 to 50 years of age) growing on sour orange rootstocks in light sandy soil of San Gabriel Valley, California. General appearance of an affected tree is similar to that caused by girdling. First symptom noticeable in top is dull, ashen color, and curling of leaves lengthwise and upward. Leaves may gradually drop in chronic cases, or suddenly wilt and dry in place on tree in acute cases of collapse. Death of fibrous roots at periphery of root system precedes top symptoms and progresses inward toward trunk, involving fibrous and larger roots. In chronic form, dropping of leaves and twig dieback keep pace with root destruction, although trees may put out new, feeble growth of foliage and roots, and continue to live. Starch disappears in affected roots and trunk, but reappears in limited amounts following new leaf growth. There is also less sugar and slower respiratory and catalase activity in affected than in healthy trees. Sieve-tube degeneration is invariably found in the sour orange stock of mature declining trees. Histological symptoms resemble those found in sweet cherries growing on Mahaleb roots and affected by Green Valley Buckskin virus.

Late wilt of flax. KOMMEDAHL, T., and J. J. CHRISTENSEN. During the last decade, several thousand lines and varieties of flax were tested annually for wilt resistance on flax-sick soil. Varieties that differ greatly in susceptibility differ also in time of wilting. Newland and Punjab tend to wilt as seedlings; Linota and C.I. 423 tend to wilt late in the season; others, like Pale Pink, Redwing, and Crystal, are variable. In general, progenies from crosses involving Newland tend to wilt early; whereas progenies containing C.I. 423 tend to wilt late. These tendencies, probably genetic, are influenced by environment, especially temperature. Punjab, wilting as seedlings in most seasons, wilted late in the cool season of 1945. The fact that varieties wilt at different times during a season is significant in varietal and inheritance studies. In late wilt, partial infection of the plant sometimes occurs. *Fusarium lini* ordinarily may be isolated from both susceptible and resistant plants. The fungus may be present internally long before wilting occurs; but the entire vascular system must be invaded before the plant succumbs. In partial wilt one side of the stem may be brown, while the other side remains green; but *F. lini* may be isolated only from the vascular bundle of the discolored side.

Seed transmission and suggested control measures for stripe smut of timothy. KREITLOW, K. W. Seed harvested from smutted plants of timothy yielded 5.8 per cent infected plants among the 877 seedlings observed. The infection originates from chlamydospores borne on the surface of the seed. When 550 contaminated seeds were surface-sterilized in 1: 500 mercury bichloride solution, no smutted plants developed. Among the 500 control seedlings, 3.5 per cent were infected. When seeds obtained from smutted plants were stored 1 year at room temperature, no smutted plants developed among 343 seedlings observed. Tests with dust fungicides are underway.

A mosaic disease of Mohawk potato caused by a virulent strain of the latent-mottle virus. LARSON, R. H. A virus causing a pronounced mosaic mottle in Mohawk has been found to be a severe strain of the potato latent-mottle group. The relationship was established by cross-immunity inoculations on tobacco with the potato ring-spot virus, by physical properties of plant extract *in vitro*, by serological precipitin reactions as well as precipitin-absorption reactions, and by the production of lethal streak on tomato by mixed infections with the virus of ordinary tobacco mosaic. The potato seedling 41956 is immune. Repeated aphid (*Myzus persicae* and *Macrosiphum solanifolii*) transmission tests have yielded negative results. The virus was readily transmitted artificially by plant extract to *Capsicum annuum, Cyphomandra betacea, Nicotiana rustica, N. glutinosa, Datura stramonium* var. *tatula, D. metel, Nicandra physalodes, Solanum aculeatissimum,* and *Schizanthus retusus.* Inoculated *Nicotiana tabacum* (var. Connecticut Havana No. 38 and White Burley) developed a very pronounced mottle with no evidence of ring lesions or line patterns. Graft transmission to the potato variety Epicure resulted in lethal top-necrosis whereas only a mottling and crinkling developed on the varieties President and British Queen. *Lamium hybridum* (Labiatae) developed a diffused systemic mottling when infected. (University of Wisconsin and U. S. Department of Agriculture.)

Perennial groundcherries as overwintering hosts of the potato yellow-dwarf and veinbanding viruses. LARSON, R. H. *Physalis virginiana* and *Physalis heterophylla* are common weeds in the potato fields in central Wisconsin. Many plants have been found that are dwarfed and have rolled and vaguely mottled leaves. Transmission tests with

nonviruliferous adults of the clover leafhopper (*Aceratagallia sanguinolenta*) to the potato variety Green Mountain resulted in the recovery of the yellow-dwarf virus from naturally infected plants of both species. Transmission tests with *Myzus persicae* to potato, *Nicotiana tabacum*, and *Hyoscyamus niger* also resulted in the recovery of the veinbanding virus. In the late fall, when most vegetation has been killed by frost and is dead, the common perennial *Physalis* remains green for a longer period, usually until late in November, affording important feeding plants for both leafhoppers and aphids. The late growing season also lengthens the period for the viruses to become established in the perennial roots of *Physalis*, and these thus become an important reservoir for and a means of overwintering of these 2 potato viruses. (University of Wisconsin and U. S. Department of Agriculture.)

Ethylene dibromide—a promising new soil fumigant. LEAR, BERT. Ethylene dibromide has given excellent control of the root-knot nematode, *Heterodera marioni*, in replicated greenhouse and field trials. Preliminary tests made in glazed, gallon crocks of infested soil showed that 0.2 cc. of the undiluted chemical was lethal to all forms of the nematode. For larger trials in greenhouse ground beds and field plots, the fumigant was diluted with propylene dichloride at the rate of 1 to 9 by volume. A commercial product, Dowfume W–10, found to be equally effective, contains an inexpensive naphtha fraction as the diluent in place of the propylene dichloride. Almost complete eradication of the nematode has been achieved with 4 cc. of the 10 per cent mixture applied 4 inches deep at staggered 10-inch intervals in rows 10 inches apart. Approximate cost per acre at this dosage rate is $75. The fumigant has successfully penetrated within 24 hours solid, sound tomato root galls in soil. Control has been equally good with and without a water seal. Its low vapor pressure causes it to persist in a sandy-loam soil, to the detriment of tomato transplants, for a period of 1 to 2 weeks at a soil temperature of 70° F. Limited trials have indicated its poor fungicidal properties.

The effect of an antibiotic substance on apple leaf infection by Venturia inaequalis. LEBEN, CURT, and G. W. KEITT. A species of *Streptomyces* was found antagonistic on agar to all of 29 phytopathogenic fungi tested, and not antagonistic to most bacteria, including those commonly used in antibiotic assays and certain phytopathogens. For the preparation of concentrates of the antibiotic substance, the antagonistic organism was grown in shake-flasks containing a corn-steep-glucose medium. The active material was obtained by ethanol extraction of the precipitate formed when the culture filtrate was acidified with HCl to pH 2.5. Further fractionation yielded a solution completely inhibiting growth of *Venturia inaequalis* at 1: 8,000,000 and *Sclerotinia fructicola* at 1: 11,000,000 (solids basis, agar-streak test method). There was no apparent loss in activity of ethanol solutions stored 11 months at 8° C. The active material is precipitated from ethanol solutions on the addition of water. In 3 greenhouse tests, infection was prevented or greatly reduced on susceptible apple leaves by a single spray application of an ethanol solution of the active material 4 hours or 4 days prior to inoculation with *V. inaequalis*.

Field resistance to leafroll infection in potato varieties. LOCKE, SETH BARTON. Twenty-three potato varieties and numbered seedlings were grown at 3 locations in Washington State in 1944 and 1945, exposing them to natural leafroll infection. The percentage of plants infected was determined by replanting samples from each plot and observing the amount of tuber-borne disease. Variations occurred in the relative performance of individual varieties at the 3 locations and during the 2 seasons, but the combined data show that the varieties form an almost continuous series with respect to amount of leafroll infection. At the most resistant end of the series are Katahdin (11 per cent) and Sequoia (18.5 per cent). The most susceptible are Netted Gem (51 per cent), Chippewa (54 per cent), and Burbank (58 per cent). Marked differences in amount of leafroll infection at the different locations are correlated with differences in recorded temperature and humidity. These weather factors appear to act through their effects upon migration of the aphid vectors.

Nutritional sprays on grapes. MACK, G. L., and N. J. SHAULIS. The addition of urea to foliage sprays as a means of controlling plant nutrition was investigated. Grape foliage was sensitive to urea, being severely injured by a single application of 4 lb. per 100 gal. The injury is of 2 distinct types, depending on whether the spray is applied to the upper or lower side of the leaf. Urea applied only to the upper surface caused marginal necrosis. Urea applied only to the under surface caused ¼-inch rounded lesions distributed over the whole surface. Copper in the form of Bordeaux mixture (3–3–100) prevented injury from urea up to a concentration of 4 lb. per 100 gal. Higher concentrations of 8 and 12 lb. urea in Bordeaux caused proportionally greater injury within 5 days. At harvest, leaves sprayed with 4, 8, and 12 lb. urea were progressively greener

than those sprayed with Bordeaux alone. Analysis of the blade for total nitrogen showed no significant differences, indicating that nitrogen from foliage sprays is not accumulated in the leaf.

Spraying and dusting rutabagas to prevent water-core (boron deficiency). MACLACHLAN, J. D. Water-core (boron deficiency) of rutabagas can be prevented by a single foliage application of borax, as a spray or as a dust, when the rutabaga root is 1 to 1½ inches in diameter. The spray contains 12 lb. of fine borax, 3 lb. of bentonite clay and ½ cup (or less) of Orthex per 40 gal., Imperial Measure. A pressure sprayer with 3 nozzles per row, applying 40 to 50 gal. per acre, is recommended. The dust contains fine borax (about 300 mesh) plus the carrier Celite, mixed in equal proportions by weight; the application of 40 to 50 lb. of this mixture per acre is recommended. Experimental results have been substantiated through commercial application by growers. Spraying and dusting are now recommended in those districts of Ontario where soil applications of borax fail to give results.

Differentiation of cultural types of Sclerotinia spp. by means of hydrogen-ion concentration. MADER, E. O., and M. N. TELLER. Sixty-two isolates of *Sclerotinia spp.*, which appeared identical on nonacidified potato-dextrose agar (pH 5.6–6.0), could be classified into 12 distinct cultural groups when grown on potato-dextrose agar adjusted to a pH range of 3.1–3.7 with additions of lactic, tartaric, malic, or citric acid, or Sorensen's sodium citrate-hydrochloric acid buffer solution. Monosporous isolates from a single apothecium and even from a single ascus did not necessarily fall within the same cultural type. The cultural types obtained from 5 ascospores isolated from a single ascus fell into three cultural groups. When representatives of the various cultural types were tested again after 8 and 12 months, they fell into the same cultural groups, although topography of types varied slightly.

The pea-seed-treatment method of evaluating fungicides in the greenhouse. MCCALLAN, S. E. A. Perfection peas are treated with experimental chemical dusts on a percentage seed-weight basis, rolled and planted in 10 seed-row units in flats or greenhouse benches of naturally infested soil. Rows are replicated several times in randomized incomplete blocks or other arrangements. Observations are taken weekly for one month on emergence and stand, the difference being post-emergence damping-off. Emergence is completed in two weeks but post-emergence damping-off continues with further time. In general, there is no evidence that seed-protectant fungicides control post-emergence damping-off expressed on absolute basis of seeds planted. The regression of height of seedlings on percentage of stand is highly significant. Dosage response curves are very flat. Increasing depth of planting delays emergence but has little final effect. Infested soil may be diluted considerably with good soil without appreciably diminishing the amount of seed decay. Used soils are composted with the pea plants, later mixed with good soil for further tests. Block effects usually are not significant. Treatment × test interaction is but little greater than replicate error. Differences of 16 per cent can be demonstrated on the basis of 50 seeds per treatment for 2 tests.

The isolation of Pythium from soil at various seasons of the year as related to soil temperature and moisture. MCLAUGHLIN, J. H. Pythium has been isolated from the surface, 3-, 6-, 9-, 12-, 15-, and 18-inch soil levels at all seasons of the year. Isolations were made at approximately weekly intervals by placing minute quantities of soil beneath 2 per cent water agar in Petri plates. Soil-temperature and moisture data were taken concurrently. The percentages of Pythium isolates were generally high in the winter, spring, and fall seasons and low in the summer. A combination of high soil temperature with low moisture generally resulted in a reduction in the percentage of Pythium isolates. Although the percentage of Pythium isolates obtained under some environmental conditions reached very low levels, seedling tests showed that *Pythium* was not absent from the soil. The corelationships between soil temperature, moisture, and percentage of Pythium isolates were distinct and consistent to such a degree that the latter could be estimated with considerable accuracy from determinations of only the first 2 variables.

Factors influencing sporulation of Sclerotinia fructicola, Venturia inaequalis, and Phytophthora infestans. MILLER, H. J. Factors favoring growth of a fungus in culture do not necessarily produce optimum sporulation. High yields of spores are desirable for slide-germination tests as well as for artificial inoculation of plants, both in greenhouse and in field. Knowledge of optimum conditions for spore production should be useful for interpreting epiphytotics, since it is the dissemination of spores which accounts, in large part, for the distribution of diseases in an area. *Venturia inaequalis* gave highest yield of conidia on 10 per cent malt agar at 18° C. Time required for

optimum yield was 25 days at 9° C.; 18 at 12° C.; 11 at 15° C.; 11 at 18° C. and 7 at 21° C. with a marked drop after these periods in number of spores obtainable in a water suspension. *Sclerotinia fructicola* gave higher yield on malt than on potato-dextrose agar with maximum number of conidia being produced on ten per cent malt agar. *Phytophthora infestans* produced highest number of sporangia at 18° C. in 21 days on Lima-bean agar. Addition of Brewer's yeast, yeast extract, riboflavin, thiamin chloride, and corn steep liquor in varying concentrations to potato-dextrose agar did not increase sporangial yield.

Heat treatments of sour cherry carrying yellows and necrotic ring spot. MOORE, J. DUAIN. In greenhouse studies in 1944 bud sticks from Montmorency trees affected with both yellows and necrotic ring spot were treated in a water bath at 50° C. ± 0.1°. The bud sticks were 15 cm. long and 2.7–3.0 mm. in diameter at the base. The periods chosen for treatment were from 2 to 16 minutes at 2-minute intervals and from 17 to 22 minutes at 1-minute intervals. Two bud sticks were removed from the bath at each interval, and 1 bud from each stick was budded into each of 2 Montmorency trees. Two trees were budded from sticks receiving no treatment, and 2 were left unbudded. In 1944 all budded trees, except 1 budded with untreated buds, showed necrotic ring-spot symptoms, and by the spring of 1946 all budded trees, except 4 that died, showed yellows symptoms. The nonbudded trees remained healthy. In a limited experiment with potted trees subjected at bud-break to heat treatment of 35° C. for 11, 19, 28, and 35 days, respectively, yellows symptoms appeared the following year on diseased trees treated for 11, 19, and 28 days, respectively, while the tree treated 35 days died. All control trees remained healthy.

Temperature and seasonal development of host in relation to expression of leaf symptoms of sour cherry yellows. MOORE, J. DUAIN, and G. W. KEITT. Eighteen potted Montmorency trees in the greenhouse were budded in 1944 shortly after bud-break with buds from a Montmorency tree affected by yellows. Before budding, all trees were incubated at approximately constant temperature of 24° C. for 9 days. Immediately after budding, 3 trees were placed at appromixately constant temperature of 16° C. Twelve were transferred in groups of 3 trees each to 16° C. after additional periods at 24° C., as follows: (1) 1 week after budding (full bloom); (2) 2 weeks after budding (petal-fall); (3) 3 weeks after budding; and (4) 4 weeks after budding. Three trees were kept at 24° C. No yellows symptoms occurred in 1944. All trees were carried out-of-doors through the summers of 1944 and 1945. In the greenhouse seasons of 1945 and 1946, all trees were forced to break dormancy at 24° C. and each tree was handled in the same manner as in 1944. Both in 1945 and 1946, abundant leaf symptoms of sour cherry yellows developed on all trees transferred to 16° C. shortly after bud-break and on those transferred in bloom. In 1945 no other trees showed yellows symptoms, but in 1946, 1 tree transferred at petal-fall had 2 yellows leaves.

A technique for making serial dilutions. MOORE, M. B., and T. KOMMEDAHL. A hypodermic syringe fitted with a 6-inch delivery tube is used to dispense accurately and aseptically known volumes of a medium to culture tubes in a dilution series. Using a sterile syringe and sterile, plugged culture tubes, a known volume of medium, say 10 cc., is added aseptically to each tube of a series. Ten cc. of a stock solution, such as a fungicide, are then added to the first tube of the dilution series. Thorough mixing is insured by manipulating the plunger of the syringe several times, while the delivery tube is inserted in the culture tube. A 10-cc. portion of the first dilution is then drawn up, delivered into the second tube, and mixed. Similar dilutions continue throughout the series. Each tube of such a series is one-half the concentration of the preceding one. Other ratios, like 1: 3: 9, 1: 4: 16, and 1: 10: 100, may be used. In 890 tubes of serial dilutions only 5 contaminations occurred. Simplicity and ease of manipulation are merits of the technique.

Production of mint species hybrids resistant to verticillium wilt. NELSON, RAY. Verticillium wilt of peppermint and spearmint is very destructive in Michigan, occurs in Indiana and is now appearing in Oregon and Washington plantings of peppermint. It has caused the abandonment of thousands of acres of the best mint soil in Michigan and has driven the industry from areas where the highest quality oil was formerly produced. Peppermint (*Mentha piperita*) is a complex hybrid, is almost completely sterile, and flowers normally 3–4 weeks later than the spearmints (*Mentha spicata*). Beginning in 1942, methods have been developed for producing species hybrids, using a noncommercial variety of *Mentha spicata* as the pollen parent. This plant is highly resistant to wilt and, although a hybrid, produces good pollen abundantly. All attempts at hybridization by hand-pollinations failed. Utilizing the positive photoperiodic response of *Mentha piperita* to supplementary illumination, delayed plantings and other methods to delay

flowering of the pollen parent, mass insect pollinations have resulted in the production of species hybrids highly resistant to wilt. The hybrids are of diverse types; the most desirable ones mature early and are highly productive of oils of good quality.

The specific pathogenesis of the Verticillium that causes wilt of peppermint. NELSON, RAY. *Verticillium albo-atrum* is reported as the cause of wilt in many different suscepts, including ornamental, fruit, vegetable, and fiber plants, ornamental and fruit trees and shrubs. Cross inoculations with isolates from unrelated host plants are usually successful. The fungus is thus characterized by a nonspecific pathogenesis. *Verticillium dahliae* has been isolated from a number of unrelated plants and successful cross inoculations have been reported. The fungus that causes the destructive wilt of peppermint is morphologically similar to *Verticillium dahliae*. In soils at maximum levels of natural field infestation and in those artificially infested with very virulent isolates and maintained at temperature and moisture levels most favorable for wilt it has not infected plants highly susceptible to isolates of *Verticillium albo-atrum*. Conversely, isolates of *Verticillium albo-atrum* from cotton, pepper, okra, eggplant, snapdragon, chrysanthemum, blackberry, maple, and other very susceptible plants have not infected peppermint under the most favorable environal conditions for wilt. The specific pathogenesis of the mint fungus for plants in the genus *Mentha* and very closely related genera, indicates the existence of dissimilar strains within the *Verticillium dahliae* group.

Effect of dust composition on adherence of copper dusts. NIKITIN, A. A. It was found that chemical composition, structure of diluents, and their rate of oil sorption have profound effect upon adherence of copper dusts treated with oil. In studying the improvement of adherence and delivery of dust it was found necessary to determine the effect of oil content of the dust. In the method of application, pressure and distance from target were of special interest. Copper dusts, containing Pyrax, Loomkill, and Eastern Magnesia, were subjected to adherence tests by varying oil content, pressure, and distance from target. Pyrax, containing 2 per cent oil, gave better adherence than Eastern Magnesia and Loomkill upon decreasing distance from target and reducing pressure. However, Pyrax, containing 1 per cent oil, gave better adherence upon increasing distance from target and reducing pressure. Eastern Magnesia, containing 1 and 2 per cent oil, gave better adherence upon increasing pressure and reducing distance from target. Substantial improvement in adherence was secured with Loomkill, containing both 1 and 2 per cent oil, upon increasing pressure and reducing distance from target.

Actinomyces and bacteria antagonistic to Actinomyces scabies. ORELLANA, RODRIGO. The growth of *Actinomyces scabies*, the cause of potato scab, is influenced by the antibiotic effect of other soil actinomyces and soil bacteria. On potato-dextrose agar plus peptone, at pH 7.0, the growth of *Actinomyces scabies* was suppressed by *Actinomyces griseus* (obtained originally from S. A. Waksman) by an isolate of the *griseus* type obtained from Waukegan silt loam, by a blue-pigmented *Actinomyces*, by an isolate from lime-treated soil, by three isolates from sulphur-treated soil, and by one isolate from manured soil. All these isolates, except *Actinomyces griseus*, were obtained at University Farm, St. Paul, Minnesota. *Actinomyces scabies* also was suppressed by *Bacillus mesentericus* obtained from potato-scab lesions at St. Paul, and 4 isolates of *Pseudomonas spp.* (soil isolates obtained from Dr. C. E. Skinner of the Department of Bacteriology, University of Minnesota). When isolates of *Actinomyces scabies* from a single potato-scab pustule were grown together in culture, some isolates were antibiotic to others.

Soil and foliage applications of nitrogen in relation to apple-scab control. PALMITER, D. H. McIntosh apple trees that received annual soil applications of Uramon (urea) increased in both yield and susceptibility to scab infection as the rate of application was increased. Plots that received no nitrogen had the lowest yields and the least scab. Trees in plots that received only foliage applications in the form of Uramon 5–100 in combination with sulphur and arsenate of lead in the petal-fall and two subsequent sprays maintained yields comparable to those of the soil-treated plots and yet were more resistant to scab. For example, in 1943 plots sprayed with flotation sulphur paste 10–100 averaged 6, 8, 11, and 13 per cent fruit scab on trees that received soil applications of 0, 2½, 5, and 7½ lb. Uramon, respectively, compared with 7 per cent infection on trees sprayed with Uramon. Similar data were obtained in 1944 and 1945. The differences in scab control in the various plots were more pronounced when less effective fungicides were used. The probable explanation for the reduced susceptibility to scab in the plots that received foliage applications of nitrogen rests in the fact that these trees were at a lower nitrogen level during the early part of the season, which was the most critical period for scab control.

Epidemiology of rust in western Canada as influenced by the introduction of stem-rust-resistant varieties. PETURSON, B. Stationary slide exposures have been made at 3 places in Manitoba each summer since 1925. These show that there has been a considerable reduction during the early part of the season, and a very marked reduction during the latter part of the season, in the numbers of stem-rust spores in the air over Manitoba, as compared with the numbers present in corresponding periods during several years prior to 1939, when susceptible varieties were generally grown. Owing, no doubt, to the fact that some of the stem-rust-resistant varieties thus far released are susceptible to leaf-rust, there has been no marked change, since their introduction, in the number of leaf-rust spores caught on slides. As a result of this reduction in stem-rust inoculum the northward and westward spread of stem rust, as well as the severity of infection, as indicated by infection on susceptible varieties in rust nurseries in western Canada, has been greatly restricted. The growing of resistant varieties has produced no change in the physiologic races of wheat stem rust occurring in western Canada. However, certain formerly rare races of oat-stem rust, to which the new resistant oat varieties are susceptible, have greatly increased in prevalence, whereas the formerly predominant races have decreased.

The effect of leaf rust (Puccinia triticina) on the yield, grade, and quality of wheat. PETURSON, B., MARGARET NEWTON, and A. G. O. WHITESIDE. In field tests carried out at Winnipeg in 1944, 1945, and 1946, to determine the effect of leaf rust (*Puccinia triticina* Erikss.) on yield, grade, and quality, leaf-rust infection ranged from an average of 22 per cent on some varieties in the rusted plots to 87 per cent on others, while practically no rust developed in the check plots that were protected from rust by applications of sulphur dust. Heavy infection reduced yield of seed by as much as 40 per cent, kernel weight by as much as 27 per cent, and bushel weight by from 1.4 to 3.5 pounds. Infection ranging upwards of 75 per cent reduced grades by one commercial grade, while light to moderate infection caused no grade reduction. The 1944 and 1945 samples only have been subjected to quality tests. These showed that leaf rust reduced the protein content of the seed of all the varieties tested. The reductions in seed protein ranged from 0.73 per cent to 1.50 per cent, and were statistically significant. Although lower in seed protein, the rusted samples generally produced loaves of greater volume than corresponding nonrusted samples. Apparently the rust, in some as yet unexplained manner, improved the quality of the gluten of the rusted samples. Leaf rust generally reduced flour yield slightly and increased carotene content.

Control of aster yellows in carrots by control of Macrosteles divisus with DDT. POUND, GLENN S., and R. KEITH CHAPMAN. In recent years carrot fields in southeastern Wisconsin have been heavily infected with the aster yellows virus, often as high as 75 per cent. In 1946 wettable DDT spray (2 lb. per 100 gal.) was applied on a commercial field at 100 gal. per acre and 400 lb. pressure. Six spraying dates were selected at 10-day intervals, beginning when carrots were 4 to 6 inches high. Treatments consisted of the following schedules of spraying dates: (A) all 6 dates; (B) first, third, and fifth dates; (C) first, second, and third dates; (D) fourth, fifth, and sixth dates; (E) first, second, and sixth dates; (F) nonsprayed check. No significant difference was found in amount of disease or in yield between treatments A, B, C, and E. In each the percentage of diseased plants was significantly less, and the yield significantly greater than those of treatments D and F. The differences in disease and yield of D and F were not significant. In treatment A, yellows was 38 per cent less and yield 4 tons per acre greater than treatment F. Vector counts made after each application showed that disease reduction was in proportion to insect control.

Lack of correlation between yield and disease control with fungicides. RICHARDS, M. C. Organic and inorganic fungicides were applied to tomato and potato plants to control Alternaria blight. The highest yields of marketable fruits or tubers were not always obtained from the plots showing the least defoliation. The fungicides are listed in descending order of disease control, and numbers in parentheses give the rating with respect to yield of marketable tomatoes or potato tubers. The asterisk indicates the addition of 2 lb. 50 per cent DDT. On tomatoes in 1946 the order was: 1. Bordeaux mixture, 6–6–100 (7); 2. Zinc dithane (zinc ethylene bisdithiocarbamate), 2–100 (2); 3. Tribasic copper sulphate, 3–100 (6); 4. Phygon (2,3-dichloro-1,4-naphthoquinone), 2–100 (4); 5. Zerlate (zinc dimethyldithiocarbamate), 2–100 (3); 6. Manganese dithane (manganese ethylene bisdithiocarbamate), 2–100 (5); 7. Fermate (ferric dimethyldithiocarbamate), 2–100 (1); 8. Check (8). On Green Mountain potatoes the order was: 1. Zerlate, 2–100* (3); 2. Zinc dithane, 2–100* (3); 3. Bordeaux mixture, 10–10–100* (5); 4. Bordeaux mixture, 10–5–100* (4); 5. Phygon, 2–100* (10); 6. Tribasic copper sulphate, 3–100* (6); 7. Manganese dithane, 2–100* (1); 8. DDT (dichlorodiphenyltrichloro-ethane), 2–100 (7); 9. Calcium arsenate, 4–100 (9); 10. Check (8). On Irish

Cobbler potatoes the order was: 1. Zinc dithane, 2–100* (3); 2. Bordeaux mixture, 10–5–100* (7); 3. Bordeaux mixture, 10–10–100* (4); 4. Phygon, 2–100* (5); 5. Tribasic copper sulphate, 3–100* (1); 6. Zerlate, 2–100* (6); 7. Manganese dithane, 2–100* (2); 8. DDT, 2–100 (8); 9. Calcium arsenate, 4–100 (9); 10. Check (10). The lack of correlation between yield and disease control occurred with both the organic and inorganic fungicides.

The influence of temperature and humidity on the development of white pine blister rust on Ribes leaves. RIKER, A. J., T. F. KOUBA, and B. W. HENRY. The influence of environment on infection of *Ribes* by *Cronartium ribicola* has been studied both in the greenhouse and at the Wisconsin blister-rust nursery, where white pines are being selected for rust resistance. Readings were made at intervals on the percentage of Ribes leaf area covered by uredia, necrosis, telia, and total infection. Uredia developed on *Ribes nigrum* at constant temperatures of 16°, 20°, 24°, and 28° C. With increasing temperatures, incubation time was shorter; area involved was larger; subsequent leaf necrosis was greater. However, telia developed as follows: abundant at 16°, good at 20°, occasional at 24°, and none at 28° C. The amount of infection on leaves, kept in a fog for 5 to 12 hours, increased progressively with time. The amount of infection on *R. cynosbati*, *R. missouriense*, and *R. nigrum* in the nursery and on *R. americanum*, *R. glandulosum*, *R. hirtellum*, and *R. triste* nearby was observed in detail during the seasons of 1942, 1943, 1944, and 1945. The development of rust in the nursery correlated closely with that at corresponding temperatures in the greenhouse. These results help to clarify the observations (1) that telia seldom develop well on most species during hot weather, and (2) that hot weather improves the chances for white pine to escape infection. (University of Wisconsin, Wisconsin Conservation Department, and United States Department of Agriculture cooperating.)

Evidence of fusion bodies from urediospore germ tubes of cereal rusts. RODENHISER, H. A., and ANNIE M. HURD-KARRER. Germ tubes from urediospores of *Puccinia graminis tritici*, *P. triticina*, *P. hordei* (*simplex*), *P. dispersa*, *P. coronata*, and *P. sorghi* growing on agar containing mineral nutrients and glucose in darkness at 10° to 25° C. have been observed to form what seem to be fusion bodies. The tip of a germ tube, growing aerially, becomes suddenly distended into a spherical body and the hyphal contents crowd into it. When first formed, it is approximately the size of the urediospore. The supporting hypha soon becomes flaccid, bends, and the fusion body may contact another germ tube. There was some evidence of a breakdown in the germ-tube wall at the point of contact, and in one instance the movement of the contents of the contacted germ tube into the fusion body was observed. These bodies in a later stage occasionally produced as many as 5 hyphae, which generally failed to develop beyond about 50 μ. The number of fusion bodies increased with increasing concentrations of glucose up to 6 or 8 per cent. Formation of the bodies was completely inhibited behind a blue filter that transmitted wave lengths of from 4000 to 5400 Å units, whereas behind red and yellow filters the bodies formed as in darkness.

Helminthosporium blight of oats in Arkansas. ROSEN, H. R. For several years an Helminthosporium disease of oats has been noted and studied which involves seedling blight, blighting of tillers of varying age, and localized blade and sheath spots. Under field conditions the disease up to the present has been noted only on varieties with Victoria parentage, while under artificial conditions in the greenhouse the disease has been produced not only on Victoria derivatives but also on a few varieties that have no such parentage. While spore measurements seemingly do not permit sharp differentiation of this pathogen from *H. avenae*, symptoms and varietal range differ markedly. Seed treatment with ethyl mercury phosphate preparations gives at least partial control of seedling blight initiated by infected seed, but poor or no control of seedling blight originated by soil-borne inoculum.

New pathogenic races of Cercospora oryzae affecting rice. RYKER, T. C. The control of *Cercospora* leaf spot, the most serious disease of rice in Louisiana, has been sought through the breeding of resistant strains. Of the mid-season and late varieties, Blue Rose has been classified as susceptible; and Rexoro, Fortuna, and Nira as resistant. Race 4, affecting only Fortuna, was reported in 1940. This race never built up to any appreciable degree. However, in 1945 several diseased fields of Fortuna were observed and a still greater number were observed in 1946. Isolates, when tested on the host differentials, indicated that a new race (Race 7) was involved, since, in addition to Fortuna, Blue Rose also was susceptible. Rexoro has become badly diseased, since 1944, with a widespread epiphytotic of Race 6. Nira, the only remaining resistant variety, showed some infection in 1945 and several severely infested fields were observed in 1946. Isolates, when tested,

showed that a new race had appeared and it is designated as Race 8. Of the 6 differential varieties used, Nira and Fortuna were susceptible, while Blue Rose, Blue Rose 41, Caloro, and Rexoro were resistant.

Source of seed potatoes and varied dosage of disinfectant in relation to control of seed-borne scab. SAMSON, R. W. Disinfected portions of 10 different lots of seed potatoes from mineral soils produced no greater percentages of scab-free and marketable potatoes than did nontreated portions of the same lots, when planted in Indiana muck soil. In direct comparison, treated portions of 15 different lots of seed potatoes originating from Indiana muck soils consistently produced higher percentages of scab-free and marketable potatoes than the corresponding nontreated portions, with wide differences among seed lots. Disinfection of the muck-grown seed potatoes apparently was not complete, for they yielded lower percentages of scab-free and marketable tubers than did either the treated or nontreated mineral-soil stocks. Light and heavy dosages of sulphur applied to slightly scabby tubers of the variety Sequoia from Indiana muck resulted in yields of 42 and 77 per cent, respectively, of scab-free potatoes when grown in mineral soil. The nontreated check produced only 11 per cent scab-free tubers. Variable responses to treatment for seed-borne scab may be due to variations in scab inoculum on different lots of seed potatoes.

The influence of climate on four leaf parasites of Zea mays in Guatemala. SEMENIUK, G., and J. R. WALLIN. Corns from the lowlands, highlands, and mountainous regions of Guatemala were planted at 6 locations in Guatemala, from sea level to 8200 ft. altitude. The climate at these locations ranged from tropical to temperate, rainfall from 40 to 150 inches, and corn maturity from 3 to 9 months. Corn rust (*Puccinia sorghi*) was the most prevalent severe disease at Tiquisate (100 ft.), Barcena (5000 ft.), Antigua (4900 ft.) and Quezaltenango (8200 ft.). *Helminthosporium turcicum* leaf blight was most prevalent and severe at Coban (4200 ft.) and Barcena. *Angiospora zeae* was prevalent in moderate amounts at Chocola (3200 ft.) and in trace amounts at Coban, Barcena, and Antigua. *Phyllachora zeae* occurred primarily on the earlier maturing sorts in slight amounts at Chocola and Coban and as traces at Barcena. The corns from the mountain regions maintained their resistance to corn rust at Quezaltenango, Barcena, and Antigua but failed at Tiquisate. Late highland corns were heavily rusted at Tiquisate, moderately at Quezaltenango, slightly at Barcena and Antigua. The early-maturing lowland corns were severely rusted only at Quezaltenango, Antigua, and Tiquisate, and moderately at Barcena. Climate influenced the prevalence and destructiveness of the leaf parasites in Guatemala. Selected locations served to uncover resistance. Two corns proved highly resistant to *Helminthosporium turcicum*. (Iowa State College and Guatemala Tropical Research Center.)

Root-necrosis resistance in maize. SEMENIUK, G., J. R. WALLIN, and I. E. MELHUS. Collections of United States, Mexican, Guatemalan, and South American corns were tested for their reaction to *Pythium graminicola*, a destructive parasite of corn roots in Iowa. Altogether, 988 collections were included in 23 different trials. A standard procedure, developed from preliminary studies, was followed. In each test 40 different lots were planted in quadruplicate in steamed infested soil and in duplicate in steamed noninfested soil. Two inbred lines, R 4 and Hy, of known reaction to *P. graminicola*, were included in each trial. Twenty-one days after planting, the seedlings were removed and the soil washed from the roots. The severity of the pathogen on the seedlings was recorded in five classes based on the amount of root necrosis and root development and reduction in seedling height. Only six lots of corn, including 3 Guatemalan, 1 United States, and 2 Mexican collections, were in the most resistant class. Seventy-one lots, including 55 Guatemalan, 9 United States, 4 Mexican, and 3 United States–Guatemalan single-cross corns were in the next highest class. The majority of the corns were in the markedly susceptible classes 3, 4, and 5.

Plant-disease relation and classification of Fusaria. SHERBAKOFF, C. D. Many Fusaria are pathogenic and still more are commonly associated with various diseases. They exhibit a great variety of forms and occur universally on all sorts of substrata. Thus plant pathologists are often concerned in their identity and would welcome an adequate classification. The author's studies support Wollenweber's classification, with these reservations: Parasitic Fusaria to retain their original binomials, such as *F. lycopersici*, and the "form" to be used only for physiologic differences. Other Fusaria should be identified by their macroconidia in optimal development, together with characters of microconidia, conidiophores, chlamydospores, and others. No single character alone could hold throughout in any classification, and no overlapping of its extremes makes a character invalid. Observing these rules no one could possibly contrive such name as *F. roseum* f.

cerealis Snyder and Hansen. The great variability of Fusaria is more imaginative than real, although, like other organisms, they are affected greatly by environment. It must be kept in mind, however, that the value of the differences between some of the Fusaria may be on a par with that between varieties of roses or wheat. Knowledge of how to obtain optimal sporulation, and good photographs, will greatly improve the classification.

New methods of artificial seed inoculation for testing the resistance of spring and winter barleys against stripe, Helminthosporium gramineum. TAPKE, V. F. The writer has recently described the "mycelial-fragments" method of inoculating barley seed with the stripe fungus, *Helminthosporium gramineum*. The method has proved consistently effective and practical to apply. In further study it has been found that after the seed has been inoculated with mycelial fragments in aqueous suspension, the duration of incubating the seed at 7°–10° C. plays a critical rôle. With increase in length of incubation the severity of stripe increases but the percentage of seed germination decreases. This decrease is due partly to the fungus and partly to the increasing injury associated with the drying of seed that has developed longer sprouts in the longer periods of incubation. In general an incubation of 4 to 6 days seems best for a heavy stripe attack with only a small to moderate reduction in seedling emergence. In studies on the stripe resistance of winter barleys grown in the greenhouse a modification of the mycelial-fragments method plus seed vernalization has given both excellent infection and heading of the winter barley plants.

Cumulative benefits from the control of leaf spot of sour cherry in West Virginia. TAYLOR, CARLTON F. A cumulative spray experiment, started in 1939 on 2-year Montmorency cherry trees, has been continued through 1946. Over this period control of leaf spot (*Coccomyces hiemalis*) with Copper Hydro 40 or with Bordeaux mixture has been much superior to that obtained with all-season lime-sulphur schedules. In 1945 heavy early defoliation on the lime-sulphur-sprayed plots had stimulated the production of secondary growth on 64 per cent of the terminals by July 21. These secondary leaves were soon lost due to leaf spot, and some tertiary growth developed. Following this poor control of leaf spot an estimated 72 per cent of the branches on the lime-sulphur plots were killed by winter injury. Four applications of either Bordeaux or Copper Hydro afforded sufficient protection to avoid "second growth" and no winter injury ensued. Fruit yields in 1946 were 0.25, 28.3, and 43.0 lb. per tree, respectively, on the lime-sulphur, Bordeaux, and Copper Hydro series. Bordeaux-sprayed fruits were smaller than those sprayed with Copper Hydro (138.8 vs. 121.8 fruits per lb.). Trunk diameter increases have been significantly greater following copper sprays than following lime-sulphur.

Changes in pectin produced by isolates of Sclerotinia spp. TELLER, M. N., and E. O. MADER. Three isolates of *Sclerotinia spp.*, which were weakly, moderately, or strongly virulent as judged by the rate at which they rotted apples, were cultured on 1 per cent apple pectin solution. Viscosity, pH, and total acidity were determined at 24-hour intervals for 5 consecutive days. Noninoculated 1 per cent pectin was used as control. Though the pH of the medium was not changed by the isolates, remaining between 3.4–3.6, the total acidity was increased gradually by the weakly and moderately virulent isolates, and increased rapidly by the strongly virulent isolate. The initial relative viscosity of 3.18 (distilled water = 1) was raised to 5.92 by the weakly virulent pathogen, and to 4.70 by the moderately virulent pathogen. The strongly virulent pathogen, however, lowered the relative viscosity of the medium to 1.41. The relative viscosity of the noninoculated control fluctuated very slightly. Similar viscosity values were obtained with the same isolates when this experiment was repeated six months later. There is an apparent correlation between the virulence of the fungus on apples and its effect on pectin.

Streptomycin production by Streptomyces griseus from peanut and soybean protein waste liquor. THORNBERRY, H. H. Peanut and soybean oil extracted meal fractions [(1) meal, (2) solution of protein and waste at pH 8.0, (3) protein precipitated at pH 4.5, and (4) waste liquor from protein precipitation] in varied amounts were added to a basal medium consisting of glucose 10 g., Bacto peptone 5 g., sodium chloride 5 g., distilled water 1000 ml., and H-ion concentration at pH 7.0. Production of streptomycin was by submerged culture in triplicate flasks shaken on a 2.5-inch reciprocating stroke at 100 cycles per minute and 28° C. for 3 days. Assays were by the paper-disc method. Peanut fractions 1, 2, and 3 were poor nutrients while fraction 4 at amounts equivalent to 30, 40, and 50 grams of the meal per liter gave yields as good as or better than yields with corn steep, about 100 units per ml. Soybean fraction 3 was a poor nutrient while fractions 1 at 1, 5, and 10 grams; 2 at 50 grams; and 4 at 20 and 30 grams per liter gave

production as good as or better than that with corn steep. These protein waste liquors are suitable nutrients for the production of streptomycin by submerged culture.

Preliminary studies on the nature of aversion in cultures of Diplodia zeae. ULLSTRUP, A. J. When two averting cultures of *Diplodia zeae* were grown together on potato-dextrose broth or on a synthetic medium with technical maltose as the source of carbon, and ammonium sulphate as the source of nitrogen, the filtrate of the medium, after sterilization, was toxic to either member of the averting pair, to any other culture of *D. zeae*, and to certain other species of fungi. Toxicity was evident in filtrates 6 days after seeding, and within 10–12 days after seeding filtrates were completely inhibitory to growth of *D. zeae* and a number of other fungi. When either member of an averting pair was grown singly, or a pair of compatible cultures grown together, in the media mentioned, no toxicity was expressed by the filtrate. The mycelial mat of a pair of averting cultures gained little in dry weight after 8 days of incubation, whereas the mycelial mat of a single culture or a pair of compatible cultures steadily increased in dry weight up to 18 days. toxic material in the filtrate was stable to autoclaving for 20 minutes at 15 lb. pressure. When toxic filtrates were extracted with chloroform the fungistatic material was removed by the solvent, leaving the aqueous fraction nontoxic. Extraction residues from filtrates on which single or paired compatible cultures had grown were nontoxic. (U. S. Department of Agriculture and Purdue University Agricultural Experiment Station.)

Meadow nematodes from brown root rot of tobacco. VALLEAU, W. D., and E. M. JOHNSON. Roots of several varieties of burley tobacco in the brown-root-rot plot at Lexington, Kentucky, and in 132 other tobacco plots in the same series in which tobacco makes slow growth after setting, were found heavily invaded with what apparently is the meadow nematode, *Pratylenchus pratensis*. The same nematode was present in roots of several grasses, legumes, and weeds that precede tobacco in the rotations in 117 plots. Tobacco roots from a continuous tobacco–small grain cover crop rotation and from a virgin bluegrass sod plot, in both of which tobacco (black-root-rot resistant) starts quickly, have proved, so far, free from meadow nematodes. Five tobacco varieties, 2 susceptible and 3 having some resistance to brown root rot, reacted similarly to brown root rot at Harrow, Ontario, and at Lexington, Kentucky, suggesting that the disease at both stations is caused by the same agent. The injury caused by meadow nematodes to the small roots of tobacco would seem to be sufficient to account for brown-root-rot injury.

Seed treatments for fall- and winter-sown onions. VAUGHAN, EDWARD K., and W. D. MOORE. Various fungicidal dusts have been studied as seed treatments to prevent seed decay and pre-emergence damping-off in fall- and winter-sown onions in Georgia. The materials used throughout the experiments were Arasan (tetramethyl-thiuram-disulphide), zinc oxide, Fermate (ferric dimethyldithiocarbamate), and Semesan (hydroxymercurichlorophenol). Cuprocide (red cuprous oxide) and Spergon (tetrachloro-parabenzoquinone) were used in the earlier tests, but results did not warrant their continued use. Arasan (1.5 per cent by weight) gave consistently good control throughout the tests and in 3 instances was significantly superior to any other material used. Fermate (1.5 per cent by weight) closely approached Arasan in effectiveness. Zinc oxide (2.0 per cent by weight), while fairly effective, did not give as consistently good results as Arasan and Fermate. Semesan was not effective. The effectiveness of these materials was not greatly influenced by fluctuations of soil temperature and soil moisture, or by soil type. Both Arasan and Fermate can be used over wide dilution ranges without loss of effectiveness or injury to the young seedlings.

A comparison of certain potato sprays in different localities in West Virginia. VAUGHN, J. R., and J. G. LEACH. A total of 12 different fungicides with and without DDT were tested, some in 3 localities under different environmental conditions. Among those giving best results were Bordeaux mixture, fixed copper, Dithane (disodium ethylene bisdithiocarbamate), and Dithane reaction product (He 178e, zinc ethylene bisdithiocarbamate). When used with DDT in 4- or 5-row replicated plots 60 feet long, Dithane was equal to Bordeaux, but when used on a commercial scale in one locality where late blight started early and was unusually severe, it was inferior to Bordeaux. Dithane reaction product in 1946 in small plots was better than Dithane and easier to use. Fixed copper with DDT gave slightly higher yields than any other spray tested except three exploratory chromate compounds. These were equal to fixed copper with DDT in the 1946 test under severe late blight conditions.

The use of the seedling inoculation technique for testing tomatoes for resistance to Verticillium wilt. VIRGIN, W. J., and J. C. MALOIT. The technique of seedling inocula-

tion as employed in determining resistance in tomato to *Fusarium* wilt can be applied with certain modifications in studying the resistance of tomato to *Verticillium* wilt. Tomato seedlings 3 inches high were inoculated by dipping the roots in a liquid suspension of *Verticillium albo-atrum*. To prepare the inoculum, mats of the fungus, grown on liquid media, were mixed in a Waring Blendor with water plus enough plain agar to make a thick consistency. Immediately after dipping, the seedlings were transplanted to sterile sandy loam soil in the greenhouse. The soil and air temperature was held as near 21° C. as possible throughout the test. Eight days after inoculation, wilting and yellowing of the primary leaves occurred, followed by yellowing in the secondary leaves. Internal thread-like strands of brown to black discoloration were observed in affected plants. Some of the plants died within 3 weeks after inoculation, while others having the described symptoms remained alive but were severely stunted. The organism was isolated from the stem and leaves of infected plants. Plants with no internal or external symptoms were considered tolerant or resistant.

Some phytopathological problems in Japan. WALDEE, E. L. Despite handicaps imposed by war and a feudalistic totalitarian system, Japanese phytopathologists have carried on their work with admirable success. Considerable progress has been made on the control of rice blast through resistant varieties and fungicidal sprays. Rice spraying is greatly aided by a rather effective epiphytotic forecasting service. The use of fungicidal sprays on cereals has been promoted by means of Government subsidies. Greater emphasis is now being placed on such locally destructive rice diseases as sclerotiosis, stem rot, and seedling blight. Control of the cereal rusts by resistant varieties is promising for wheat. Barley breeding is not so far advanced. Of special interest is wheat stripe (*Cephalosporium gramineum* Nishikado et Ikata) and barley bunt (*Tilletia pancicii* B. et R.). An elaborate research program on snow blight of cereals (*Typhula spp.*, *Fusarium spp.*, and *Pythium spp.*) promises to increase greatly wheat and barley production throughout the heavy snow area. The excellent research on black rot of sweet potato has received official commendation. The principal causes of low yields of Irish potatoes are virus diseases. The seed-potato certification program has not proved effective, chiefly because of inadequate enforcement of disease tolerances. The Japanese plant-disease survey is unsatisfactory and is now being reorganized.

Improvement of cabbage for disease resistance. WALKER, J. C., and GLENN S. POUND. Selection for mosaic resistance has been continued within the yellows-resistant varieties, Wisconsin All Seasons, Wisconsin Ballhead, and Wisconsin Hollander. An improved strain of Wisconsin Ballhead, released in 1946 as Improved Wisconsin Ballhead, is distinctly higher in mosaic resistance. An improved strain of Wisconsin All Seasons ready for release is distinctly higher in mosaic resistance and more uniform in desired horticultural characteristics. Selection is being continued within this variety for still greater mosaic resistance and higher ascorbic acid content. Wisconsin Hollander contains only Type B resistance to yellows and is very susceptible to mosaic. By crossing with Wisconsin Ballhead and selecting for Type A yellows resistance and mosaic resistance, homozygous Type A yellows-resistant lines distinctly higher in mosaic resistance have been secured. Selection for horticultural type is still in process.

Natural and cultural occurrence of the ascogenous stage of Diaporthe phaseolorum var. sojae. WELCH, A. W. The ascogenous stage of *Diaporthe phaseolorum* var. *sojae* has been reported only from culture. It occurs naturally during late spring, under Iowa conditions, on soybean stems overwintered in the field. Perithecia are produced singly or in caespitose groups of 1 to 7 per stroma. The perithecial beaks vary from 280 to 546 μ in length; asci 27.2–40.8 × 6.8–8.5 μ; and ascospores 8.5–10.2 × 3.4–5.1 μ. Isolates obtained from the Phomopsis stage failed to produce perithecia in culture. Isolates obtained from the stem lesions of wilting and dying plants produced perithecia in approximately 30 days on potato-dextrose agar. In culture, 1 to 32 perithecial beaks have been observed per stroma. These beaks varied from 268 to 569 μ in length; asci 33.3–51.8 × 5.6–9.0 μ; and ascospores 9.3–11.3 × 2.5–4.6 μ. Stems with lesions collected during August and September produced perithecial beaks after 4–15 days in moist chambers. Pycnidia were produced abundantly on stems of maturing plants. Pycnidia and perithecia have not been observed within the same lesion. (U. S. Department of Agriculture and Iowa Agricultural Experiment Station.)

A method of correcting for soil variation in field tests. WELLMAN, R. H., H. W. THURSTON, JR., and F. R. WHALEY. It is often impossible to distinguish between treatments because of soil variations within replicates. Statistical designs, now known to compensate for this variation, are inflexible as regards numbers of treatments and replicates. Our method has no such restrictions. Consider an experiment where plots run

north and south and are arranged side by side. Thus the field runs east and west. Yield is recorded independently for north and south halves of each plot. Long narrow plots should be further divided. Only north half plot yields are now considered. Field average plot yield ($\overline{Y}$) and average plot yield per treatment (y) is determined. R is actual yield per plot. $\overline{Y}/y \times R = G$. G values include individual variation (error) and soil variation but exclude treatment effect. Plot G values in field order. Construct a smooth curve through them by customary procedures. Subtract $\overline{Y}$ from the smooth curve at the location of each plot. The resultant value is c. $\overline{Y}/(\overline{Y} + c) \times R =$ adjusted yield. Repeat for south half. Add adjusted yields for both halves. Resultant adjusted entire plot yields are subject to analysis. In a potato experiment, the uncorrected error variance was 2840; with triple lattice correction, 2129; with above correction, 1302. (Carbide and Carbon Chemicals Corp., Pennsylvania State College, and Linde Air Products.)

The development of increased tolerance to sodium arsenite by Sclerotium rolfsii and Sclerotium delphinii. WILSON, COYT. A strain of *Sclerotium rolfsii*, 1 of *S. delphinii*, and 1 intermediate between the 2 species have increased their tolerance for sodium arsenite from 125 p.p.m. to 150 p.p.m. during 5 transfer generations covering a period of approximately 10 months. Cultures that have been on arsenical media for 5 transfer generations grow approximately twice as fast on potato-dextrose agar containing 125 p.p.m. sodium arsenite as those that have been on media containing no arsenic. No sclerotia are formed on the arsenical media, and the mycelium is brown and appressed rather than white and fluffy. When cultures that have been grown on arsenical media are returned to potato-dextrose agar they form sclerotia that are indistinguishable from those formed in cultures grown continuously on arsenic-free potato-dextrose agar. Sectoring on arsenical media is rare. Occasionally a mutant arises that does not form sclerotia.

A survey of the fungi associated with peg and seed rots of peanuts in southern Alabama. WILSON, COYT. Isolations from pegs, shells, and seeds have revealed that the young peanut is often invaded by soil-borne fungi soon after it penetrates the soil. Infection is facilitated by, but is not dependent upon, various types of insect injury. Such miscellaneous fungi as *Penicillium, Aspergillus, Trichoderma, Rhizopus*, and other Mucors have comprised more than half of the cultures obtained. Of the remainder, *Sclerotium rolfsii, Sclerotium bataticola, Diplodia theobromae, Rhizoctonia spp.*, and *Fusarium spp.* were obtained in approximately equal proportions, although in any given sample any one of these may predominate. The results indicate that *Sclerotium rolfsii* may have been overemphasized in the past as a cause of peg rot and that the disease may be caused by any of several fungi. After the peanuts are harvested, another type of seed decay, concealed damage, becomes important. In the early stages of the disease, *Diplodia theobromae* has comprised more than 80 per cent of the fungi isolated. As the decay progresses, species of *Fusarium* and other secondary invaders are obtained more frequently.

Therapeutic treatments for bean rust. YARWOOD, C. E. Destruction of bean rust (*Uromyces phaseoli typica*) uredial pustules after they were visible to the naked eye at 4 days and more after inoculation has been accomplished with little host injury by treatment with cyanide gas, hydrogen sulphide gas, vapors from dilute lime-sulphur, penetrating lime-sulphur sprays, hot water, and hot air. With lime-sulphur sprays, the addition of a spreader, treatment of the plants during the morning, the water-soaking of the leaves with sprays of high-impact pressures, and the incubation of the treated plants in dark moist chambers until after natural darkness, all favored eradication, and the optimum concentration was about 0.3 per cent lime-sulphur. Several other sprays were relatively ineffective. With hot water the time for complete killing of rust pustules without leaf-killing ranged from about 80 min. at 35° C. to 2 sec. at 55° C., with a temperature coefficient of about 50. The killing of 4- to 6-day-old pustules usually left only faint chlorotic spots on the leaf, but killing of 8- to 10-day-old pustules left necrotic areas extending roughly to the limits of the rust mycelium.

Relation of soil moisture and nutrient concentration to the development of bean powdery mildew. YARWOOD, C. E. Pinto beans, grown at low soil moisture, supported a more luxuriant growth of mildew (*Erysiphe polygoni*) with more spores formed per unit area, less leaf necrosis, and with greater reduction in host yield, than at high soil moisture. The more vigorous mildew development on slow-growing plants at low soil moisture than on fast-growing plants at high soil moisture is a marked exception to the general rule that obligate parasites grow best on vigorous growing plants. On beans grown in water cultures containing $\frac{1}{4}$, $\frac{1}{2}$, 1, 2, and 4 times the concentration of salts in standard Hoagland solution mildew development increased, especially on the primary leaves, with increasing nutrient concentration, while plant growth was best at the standard concentration. It is believed that similarities between low moisture in soil cultures and

high nutrient concentration in water cultures may explain the similarity in mildew response in these unfavorable environments for host growth, and that both situations lend further support to the idea of the xerophytic nature of many powdery mildews.

Greasy pod—a new virus disease of beans. ZAUMEYER, W. J., and H. REX THOMAS. In 1944 an apparently new mosaic was noted in much of the garden bean seed acreage of southern Idaho. In 1945 it was observed in Montana, Wyoming, and Colorado but was not so widespread in these States as in Idaho. The symptoms consist of a chlorosis and bronzing of the leaves and a shiny or greasy appearance of the pods, which are completely without pubescence and darker than normal ones. Unlike the symptoms of bean virus 1, the leaves of plants infected with greasy pod are thicker than normal with no puckering, malformation, or distortion, but are decidedly rugose. The plants are only slightly stunted, although the pods are frequently malformed because of improper ovule development. The most susceptible varieties are those that are tolerant only to bean virus 1; those resistant to greasy pod are resistant also to virus 1. In this respect greasy pod virus differs from the virus reported from New York and Idaho, which is highly infectious to some of the varieties definitely resistant to bean virus 1. The virus of greasy pod is seed-borne to about the same extent as bean virus 1. In the greenhouse the symptoms are similar to those produced by virus 1. Because of certain similarities in seed transmission, varietal resistance, and properties, greasy pod is believed to be a strain of bean virus 1.

Phytophthora cinnamomi in relation to avocado decline. ZENTMYER, GEORGE A., and L. J. KLOTZ. Evidence indicates that *Phytophthora cinnamomi* may be a primary factor in "decline" of avocado trees. This fungus is intimately associated with the disease that has killed several thousand large trees. Injury initially appears on poorly drained, wet soils. The most common underground symptom is destruction of feeder roots. The fungus has been shown capable of destroying small roots, causing cankers on larger roots and on the rootstock and trunk, even under nonwaterlogged conditions. The sudden "collapse" of avocado trees under the anaerobic conditions prevailing when excess water persists in the root zone may be entirely an oxygen relation, distinct from the slower root rot. Seedlings will "collapse" from waterlogging periods of 8 to 10 days. Seedlings in soil inoculated with *P. cinnamomi* will "decline" after two-day waterlogging periods. Treating soil from decline areas with steam or chloropicrin in the greenhouse has resulted in marked response of avocado seedlings over their growth in nontreated soil, further indicating the importance of a biological factor. Ethylene dibromide, Dowfume N (mixture of dichloropropane and dichloropropylene), chloropicrin, and 8-hydroxyquinoline benzoate, at low concentrations (0.01 per cent or less by volume) inhibit the growth of *P. cinnamomi* in soil in the laboratory. Such concentrations of ethylene dibromide and 8-hydroxyquinoline benzoate are not toxic to avocado roots.

ELECTRON MICROSCOPE STUDIES ON TOBACCO-MOSAIC VIRUS

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INTRODUCTION

In the original description of purified tobacco-mosaic virus it was reported that, on the basis of results obtained in two preliminary experiments on osmotic pressure and diffusion, the purified material appeared to possess a molecular weight of the order of a few millions (26). Since then numerous publications on the size and shape of tobacco-mosaic virus have appeared (5–21, 23, 25, 27, 28, 30). It was early recognized that the original method of preparation resulted in aggregation of the virus and since 1936 the virus has been purified in this laboratory by differential centrifugation, for this method causes little or no aggregation (1, 23, 24). Before the advent of the electron microscope it was estimated by indirect methods, based on sedimentation, diffusion, viscosity, and X-ray data, that the virus particle possessed a molecular weight of about 40 millions and was an elongated particle about 12 to 15 mμ in diameter and about 400 mμ in length (5, 16, 17). In an early study with the electron microscope the predominating unit present in centrifugally purified virus preparations was found to be a rod about 15 mμ in diameter and about 280 mμ in length (28). Similar results were obtained in further, more precise, studies involving indirect methods (20). However, in the electron micrographs, images of particles formed by the end-to-end aggregation of this unit, as well as of particles having shorter although variable lengths, were also found to be present. Lauffer (19) has shown that the sedimentation constant of the infectious principle of tobacco-mosaic virus is the same, within a probable error of 6 per cent, as that of the rod 15 by 280 mμ. His results prove that, in the case of the present material, the sole carriers of virus activity cannot possibly be particles any smaller than half the size of the predominating particles. This result is in complete accord with much earlier work from this laboratory relating virus activity directly to the nucleoprotein of high molecular weight. In the absence of data to the contrary is has seemed justifiable to conclude that tobacco-mosaic virus is actually a particle 15 by 280 mμ. It is recognized that, as with any biologically active material, the biological activity may at some future time be found to be associated with smaller particles. Perhaps with this in mind, Bawden and Pirie have, from time to time, advanced the idea that the rods 15 by 280 mμ are really aggregates and that the true virus is a much smaller, perhaps even spherical, particle (1, 2). In the absence of supporting experimental data they have been forced into the rather unusual position of "postulating that the primary virus particles are unable to exist free in solution" (3). Obviously if the virus particle cannot exist free in solution it is removed from the ordinary realm of experimentation and it becomes difficult, if not

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impossible, to prove or disprove its real existence by means of experiments. In the absence of direct experimental proof for its existence such a particle must remain a figment of the imagination. In view of the large amount of experimental data which indicates that tobacco-mosaic virus activity is directly associated with the particles 15 by 280 mμ, it would appear desirable to continue to accept the 15 by 280 mμ particle as tobacco-mosaic virus until such a time as virus activity is proved experimentally to be associated with a smaller particle.

It was found in earlier work that the characteristic rods 15 by 280 mμ could be demonstrated by means of the electron microscope in the unpurified juice pressed directly from a tobacco-mosaic-diseased plant (27). However, because of the lack of contrast of the rods with respect to residual extraneous material, the micrographs were rather unsatisfactory. The new shadow-casting technique (29) provides a method for increasing the contrast and in the present investigation excellent micrographs of unpurified infectious juice have been obtained. These show the presence of large numbers of a unit 15 by 280 mμ. In addition, in the present investigation, the rods of shorter and variable lengths that are known to be present in centrifugally purified preparations of tobacco-mosaic virus have been separated by fractional centrifugation and studied by means of the electron microscope and by means of virus-activity measurements. The results obtained provide additional reasons for considering that the rods 15 by 280 mμ occur within the plant cells and that these are, in fact, tobacco-mosaic virus.

EXPERIMENTAL WORK

In the present investigation an RCA Console Model electron microscope, type EMC-1, was used. The accelerating voltage was 30,000 volts and the magnification yielded by the microscope was 5800. The specimens were mounted on thin collodion membranes supported by copper screens. The virus samples were first diluted to a suitable degree by the addition of distilled water. Purified preparations were usually used at a concentration of 10^{-4} gm. per cc. A small drop of the diluted solution was placed on the collodion membrane and allowed to dry. In most cases the shadow-casting technique (29), involving the evaporation of gold on the screen in a vacuum at an oblique angle to the surface of the membrane, was used. In order to permit more accurate measurements of particle lengths, the image on the plate was enlarged photographically six times to yield a total magnification of 35,000.

In the first experiment young Turkish tobacco plants grown in the greenhouse were infected with tobacco-mosaic virus. Twenty-five days later the plants were harvested and frozen overnight. The frozen plants were put through a meat grinder and the juice was pressed from the mash after thawing. Electron micrographs were made of this juice directly after preparation and after standing for 1 and 20 days at 4° C. Samples of the juice were diluted with 50 volumes of distilled water immediately before examina-

tion. Figure 1 shows an electron micrograph of the juice directly after preparation and figure 2 shows the size distribution of the rod-like particles present in several pictures of this sample. All rod-like particles within given areas were included in the measurements. The total number of particles measured was 1350. It is obvious from figures 1 and 2 that most of the particles are about 280 mμ in length but that there are a few shorter as well as longer rods. Most of the longer rods fall into two groups having lengths approximately 2 and 3 times the length of the predominating unit. The very small particles forming the background for the rods are not characteristic of juice from diseased plants, for they are present in micrographs of untreated

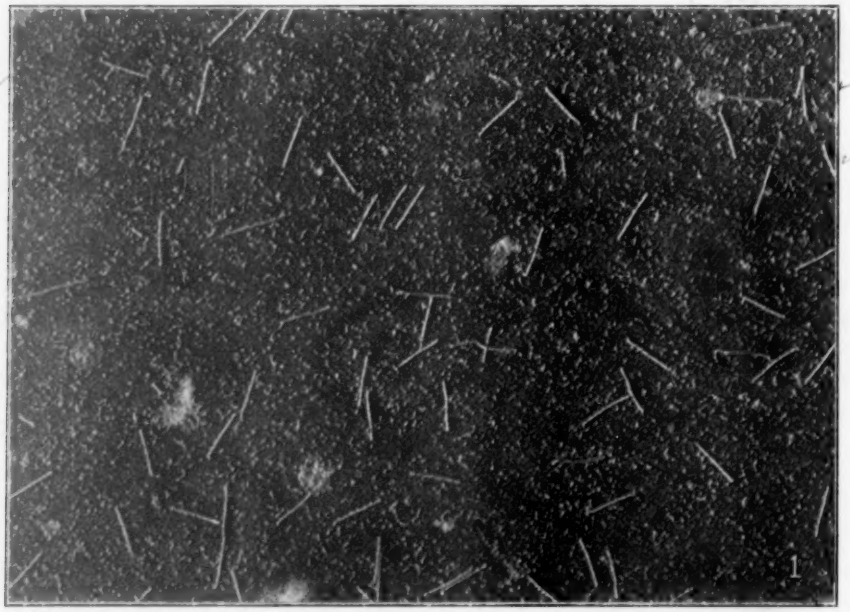

FIG. 1. Electron micrograph of the freshly expressed juice from tobacco-mosaic-diseased Turkish tobacco plants. Juice diluted with 50 volumes of distilled water and mount prepared with gold by the shadow-casting technique. The magnification is 23,200.

juice from normal Turkish tobacco plants. It is difficult to count accurately the particles of smallest size and for this reason the lower limit shown on the size distribution diagrams does not have any real significance.

Figure 3 shows the size distribution of rod-shaped particles present in micrographs taken of the juice after standing for 1 day at 4° C. It can be seen that most of the particles still fall in the size group near 280 mμ but that the group at double this size is much larger. Figure 4 shows an electron micrograph of the same juice after standing at 4° C. for 20 days. It is obvious that the proportion of rods of double length is much greater. It can be seen from the size distribution, which is shown in figure 5, that there are two major groups and that the group consisting of double length particles is even larger than the group having lengths near 280 mμ. There are, in ad-

dition, numerous particles having lengths falling between the lengths of the single and double length particles. The results indicate that as the juice is allowed to stand there is an end-to-end aggregation of the rods followed possibly by a break-up of some of the aggregates into rods of shorter and variable lengths. The virus rods shown in figure 4 appear to have a larger diameter than those in figure 1. This is due to the fact that more gold was evaporated on the specimen of figure 4 than in the case of figure 1. The amount of gold collected on the rods also depends on their angle to the beam of gold. This accounts for the different appearance of rods lying in different directions.

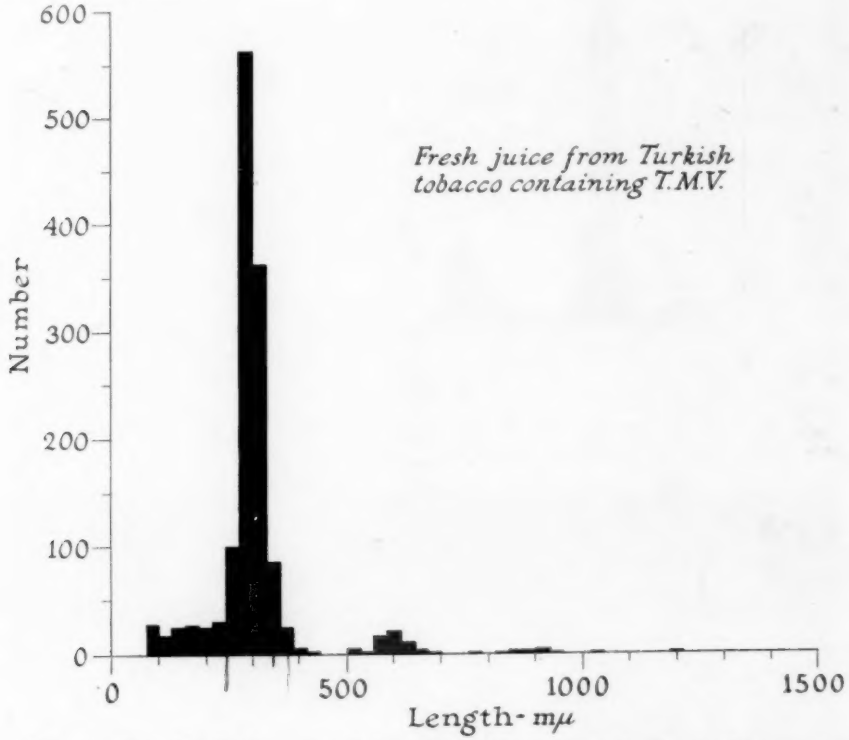

FIG. 2. Size distribution of rod-shaped particles present in the freshly expressed juice from tobacco-mosaic-diseased Turkish tobacco plants. Prepared from figure 1 and similar micrographs.

In the second experiment a study was made of the effect of centrifugal purification of the freshly expressed juice from tobacco-mosaic-diseased Turkish tobacco plants. The juice was clarified by centrifugation at about 3000 r.p.m. for 10 minutes, then passed through a thin layer of Celite and centrifuged at 24,000 r.p.m. for 1 hour. The pellets of virus were dissolved in 0.1 M phosphate buffer at pH 7 with rapid stirring by means of a mechanical stirrer. This solution was diluted with distilled water and examined. Figure 6 shows an electron micrograph of the preparation and figure 7 gives the size distribution as obtained from the particles present in

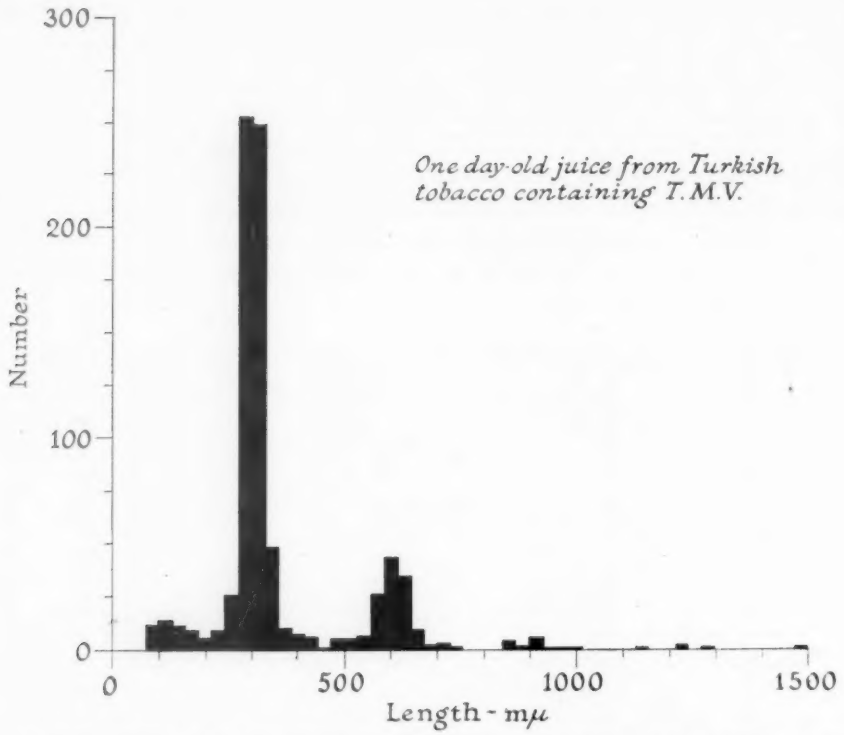

FIG. 3. Size distribution of rod-shaped particles present in the juice from tobacco-mosaic-diseased Turkish tobacco plants after standing for 1 day at 4° C.

FIG. 4. Electron micrograph of juice pressed from tobacco-mosaic-diseased Turkish tobacco plants and allowed to stand for 20 days at 4° C. The juice was then diluted with 50 volumes of distilled water. The mount was prepared with gold by the shadow-casting technique. The magnification is 23,200.

figure 6 and in similar micrographs. The preparation contains a large group of particles having lengths near 280 mμ together with a large group having shorter although variable lengths. These shorter particles are usually not found in large amounts in micrographs of virus preparations obtained by centrifugation for one hour or less at 24,000 r.p.m. followed by solution of the pellets with gentle stirring and their presence in the present preparation may be due to the vigorous mechanical stirring to which the preparation was subjected. Considerable evidence has been obtained which indicates that

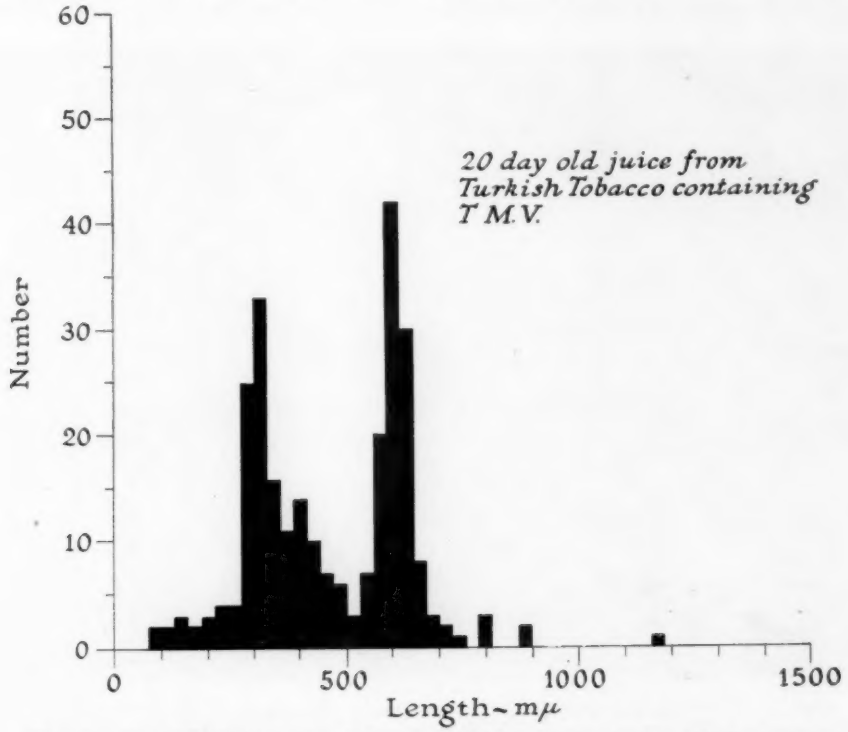

FIG. 5. Size distribution of rod-shaped particles present in the juice pressed from tobacco-mosaic-diseased Turkish tobacco plants after standing for 20 days at 4° C. Prepared from figure 4 and similar micrographs.

virus activity is associated with the rods 280 mμ in length (17, 19, 28). It seemed of interest therefore to secure an essentially pure preparation of the shorter rods so that it might be possible to determine directly their virus activity.

Accordingly, in a third experiment a comparable fairly concentrated preparation of virus was centrifuged for 2.5 hours at 24,000 r.p.m. The supernatant liquid was removed and centrifuged for 40 minutes at 24,000 r.p.m. The supernatant liquid from this centrifuge run, which was found to contain 0.15 mg. of protein per cc., was examined. It can be seen from the distribution of particle lengths shown in figure 8 that about 98 per cent of the particles are shorter than 280 mμ. The specific biological activity of this

preparation was compared with that of the original virus preparation by means of the half-leaf method on *Nicotiana glutinosa* plants (22). A solution containing 0.1 mg. of the short rods per cc. and a solution containing 0.001 mg. of the original virus preparation per cc. were used. The solution of short rods gave about 10 lesions per half-leaf, whereas the original virus preparation gave about 30 lesions per half-leaf. Since there was a hundredfold difference in concentration it is obvious that, at the same concentration, the virus activity of the preparation of short rods is less than 1 per cent that of the original virus preparation. This small residual activity is prob-

FIG. 6. Electron micrograph of tobacco-mosaic virus preparation obtained by centrifuging the freshly expressed clarified juice of tobacco-mosaic-diseased Turkish tobacco plants for 1 hour at 24,000 r.p.m. The preparation was diluted with distilled water to a concentration of 10^{-4} gm. of protein per cc. The mount was prepared with gold by the shadow-casting technique. The magnification is 23,200.

ably due to the few rods of 280 mμ length or longer still remaining in the preparation. The number of these rods is only 2 per cent of the total number of rod-like particles, but by weight this would account for about 10 per cent of the total protein content of the solution, hence a biological activity about 10 per cent of that of the original virus preparation would be expected. The fact that the activity was less than 1 per cent shows that the activity of the long rods is reduced, either intrinsically or possibly because of an inhibitory effect caused by the short particles. In any case it seems reasonable to conclude that the rods shorter than about 280 mμ are devoid of virus activity. The source of these short rods is not known with certainty. They seem to be present in the juice as it is pressed from the plant material. It is

possible that they are formed by the break-up of the virus rods due to thermal agitation. In the case of centrifugally purified virus preparations the short rods may result from mechanical stresses during centrifugation and during the subsequent solution of the virus pellets by mechanical stirring. That rods can be broken by mechanical stress is shown in figure 9, which is an electron micrograph of a mount of purified cucurbit-mosaic virus in which the collodion film has broken. The stretching of this film has resulted in the breaking of several virus rods.

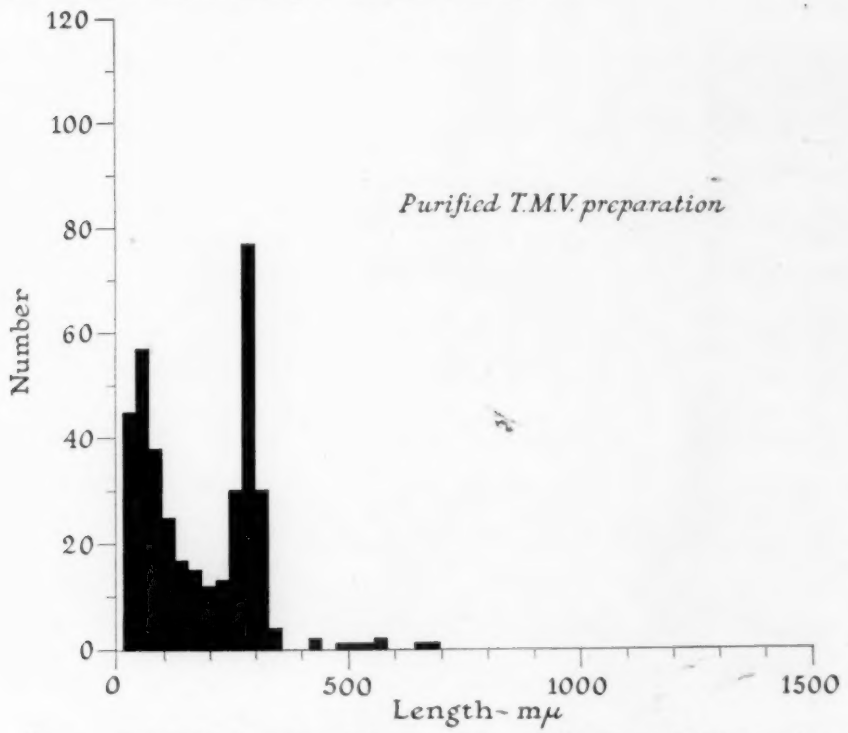

FIG. 7. Size distribution of particles present in centrifugally purified tobacco-mosaic virus preparation. Prepared from figure 6 and similar micrographs.

The cause of the end-to-end aggregation which occurs when infectious juice is allowed to stand is not known. In a purified virus sample suspended in disodium phosphate buffer at pH 7 it does not seem to take place to any appreciable extent. The juice has a pH of 5.4, but decreasing the hydrogen ion concentration by means of a buffer does not prevent the aggregation, for it takes place even at pH 9. An attempt to break up the long rods by decreasing the hydrogen ion concentration was not successful. At about pH 11.3 the virus particles are partially dissolved, as shown in figure 10, but no break-up into smaller parts seems to take place. When the hydrogen ion concentration is adjusted around or below the isoelectric point (pH 3.5), there seems to be an increase in the rate of end-to-end aggregation, but another process, the side-by-side aggregation, appears to come into prominence.

Figure 11 shows a micrograph of a sample of fresh juice which has been brought to pH 2.2 by the addition of hydrochloric acid. This picture gives a good idea of the flexibility of the virus rods. Near the middle of the picture a bundle of rods is seen to bend sharply as it crosses over a single rod lying on the collodion membrane. This side-by-side aggregation seems to be identical with beginning crystallization and as the picture is sharp enough to show the individual rods, the distances across known numbers of these can be measured. These measurements give a value of about 150 Å for the diam-

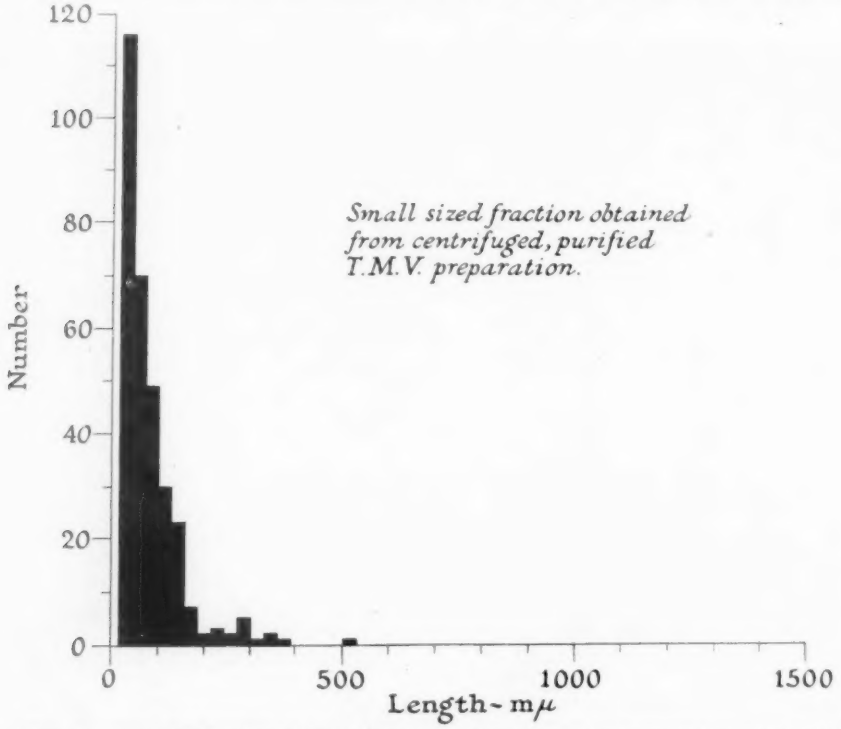

FIG. 8. Size distribution of rod-shaped particles present in preparation obtained by centrifugal fractionation of material similar to that described in figures 6 and 7.

eter of the individual rods. This value is in agreement with the value obtained by Bernal and Fankuchen (5) in X-ray studies.

DISCUSSION

The demonstration, not only that rods 15 by 280 mμ exist in the freshly expressed untreated juice of tobacco-mosaic-diseased Turkish tobacco plants, but especially that most of the rod-like particles present in such juice are essentially 15 by 280 mμ is considered of importance. If, as Bawden and Pirie (3) have postulated, the primary virus particle is small and not greatly elongated and unable to exist free in solution, it would appear most unusual for such small particles to aggregate, within the few minutes required to press out the juice, to form essentially uniform rods 15 by 280 mμ

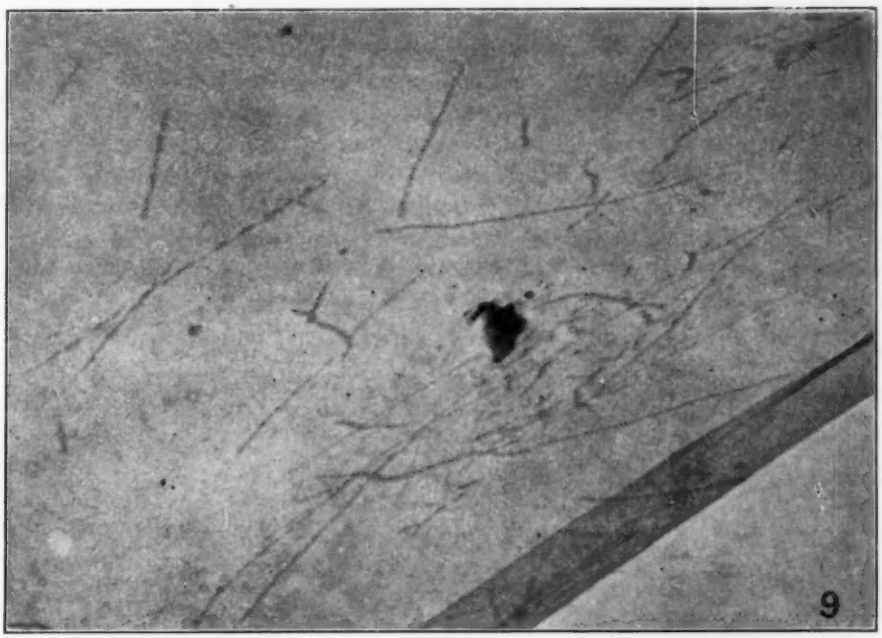

FIG. 9. Electron micrograph of cucurbit-mosaic virus particles that have been broken by the breaking and stretching of collodion membrane of mount.

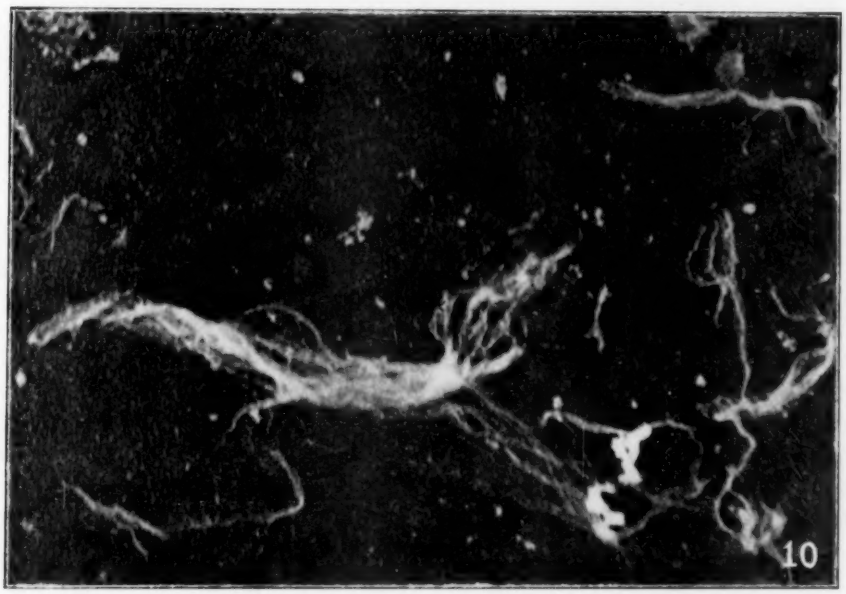

FIG. 10. Electron micrograph of tobacco-mosaic virus particles in infectious juice following exposure to pH 11.3. Mount prepared with gold by the shadow-casting technique. The magnification is 23,200.

in size. The real existence of biologically active particles 15 by 280 mμ has been amply demonstrated and it seems more logical to assume, as has been done in this laboratory for some years, that these particles represent tobacco-mosaic virus. If one wishes to postulate the existence of biologically active particles of smaller size, it should be accompanied by acceptance of the burden of proof for the demonstration of the real existence of such particles. In the present investigation, as well as in work by Bawden and Pirie, preparations of particles of smaller size have been obtained, but have been found to possess little or no virus activity. Despite the absence of virus activity Bawden and Pirie persist in referring to such material as "virus." The essence of a virus is its virus activity and it seems unwise to

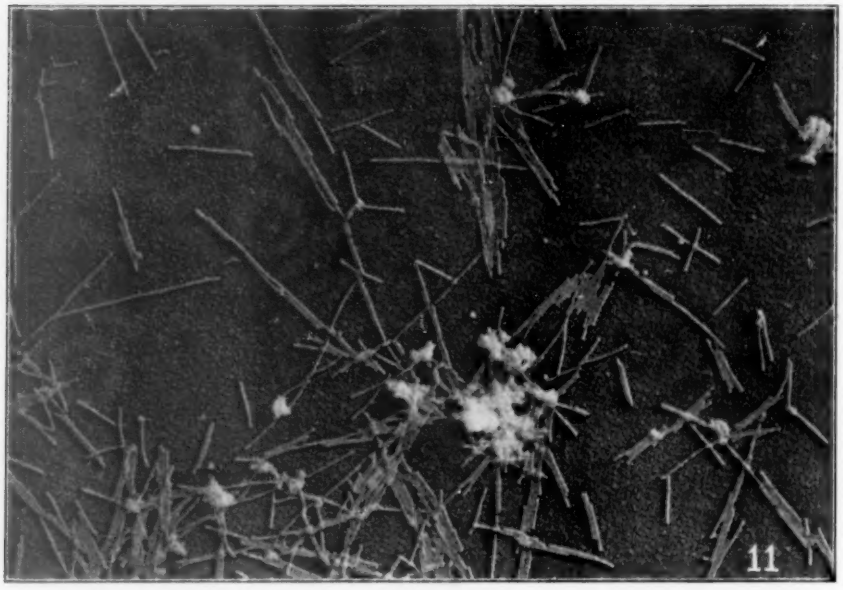

FIG. 11. Electron micrograph of tobacco-mosaic virus particles in infectious juice following adjustment to pH 2.2. The mount was prepared with gold by the shadow-casting technique. The magnification is 23,200.

refer to a material possessing no virus activity as "virus." Much experimental evidence exists which indicates that the rods 15 by 280 mμ represent the minimal biologically active units of tobacco-mosaic virus and it would appear reasonable to continue to accept this material as tobacco-mosaic virus until it can be proved experimentally that virus activity is associated with units of smaller size.

The origin and properties of the particles smaller than 15 by 280 mμ and the end-to-end aggregation of the 15 by 280 mμ units represent fields which will require considerable additional investigation. Little is known concerning the origin of the rods shorter than 280 mμ, but there seems to be general agreement that they may arise from the break-up of the longer rods as a result of thermal or mechanical stresses. There is some evidence which

indicates that the joint which is formed by the end-to-end union of two rods 280 mμ in length is just as strong mechanically as other points along the newly formed rod 560 mμ in length. Thus the end-to-end union of rods followed by the breaking up of the long rod into rods of uneven lengths could serve to explain the existence of rods of variable lengths, such as those shown in the micrographs. It has been demonstrated quite conclusively that preparations of the very short rods possess no virus activity, but the question of the virus activity of aggregates of the 15 by 280 mμ units is less clear, although there is some evidence that such aggregates possess a slightly diminished virus activity (23). It is possible that any rods that are about 280 mμ or more in length can serve as infective units, although multiples of the basic unit may be necessary. It does seem likely that the conversion of a preparation consisting essentially of rods about 280 mμ in length into a preparation consisting essentially of rods about 560 mμ in length should be accompanied by a 50 per cent reduction in specific virus activity due to the reduction in the actual number of particles present.

It will be difficult, if not impossible, to demonstrate conclusively that all of the tobacco-mosaic virus within the cells of diseased Turkish tobacco plants exists in the form of particles of a given uniform size. However, the present demonstration that most of the rod-shaped particles present in the freshly expressed juice of diseased plants are about 15 by 280 mμ in size, the isolation of most of the biologically active material in the expressed juice in the form of rods 15 by 280 mμ and Beale's work on the nature of virus within living cells (4), provide considerable justification for the assumption that tobacco-mosaic virus exists within plant cells in the form of rods 15 by 280 mμ.

SUMMARY

Electron micrographs, prepared by the shadow-casting technique, of the freshly expressed juice of tobacco-mosaic-diseased Turkish tobacco plants show that most of the rod-shaped particles present are 15 by 280 mμ in size. On standing at 4° C. many of these particles in the juice join end-to-end to form particles of greater length. At the end of one day a significant increase in the number of particles of double length was evident and at the end of 20 days the number of particles of double length was greater than the number of particles having lengths near 280 mμ. The joints formed by the end-to-end union of particles appeared to be as strong mechanically as other positions along the rods. Some of these elongated particles appear to be broken up by thermal or mechanical stresses to form particles of shorter and variable lengths. Particles less than about 280 mμ in length appear to possess no virus activity.

The results provide additional justification for the assumption that the basic infective unit of tobacco-mosaic virus is 15 by 280 mμ in size.

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THE EFFECTIVENESS OF D–D AS A SOIL FUMIGANT IN HAWAII[1]

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Numerous chemicals have been tested as soil fumigants in controlling the root-knot nematode, *Heterodera marioni* (Cornu) Goodey, and other plant pathogens in the soil. Until recently the most successful and widely used material has been chloropicrin. The high cost and difficulties of applying this fumigant have precluded its large-scale field use with most vegetable and agronomic crops. A recently discovered fumigant known as D–D[2] is much easier to apply and cheaper to produce. The purpose of this study has been to determine the effectiveness of D–D as a seedbed and field fumigant under Hawaiian conditions. Although primary emphasis has been placed on nematode control, data are presented which indicate that it may be effective against other soil-borne organisms.

REVIEW OF LITERATURE

The effectiveness of D–D as a soil fumigant was first reported by Carter (1) in 1943. Carter's results were particularly striking in an area where a complex including at least *Anomala* beetle larvae (*A. orientalis*), nematodes, and pythiaceous fungi had resulted in serious plant failure. Pineapple plants in treated areas gradually showed increasing improvement over non-treated checks. Carter concluded that, apart from its immediate effect in reducing populations of harmful organisms, it had also affected beneficially the soil complex.

Christie (4) found D–D to have an effective killing range of from 9 to 12 inches when applied at a depth of 6 inches in a Berwyn loam soil. Variation in killing range from test to test did not seem to be correlated with soil moisture or temperature. D–D was more effective against nematodes in undecomposed roots than was chloropicrin.

Parris (5) found D–D to be an effective nematocide at rates as low as 150 lb. per acre in a silt-loam soil without a soil cover other than a wetting of the surface. He was able to demonstrate very little fungicidal value from studies with damping-off fungi, species of *Rhizoctonia*, *Fusarium*, and *Pythium*. A slight phytocidal action was noted if plants were set out too soon after treatment. A delay of at least 2 weeks between treatment and planting was recommended. The time interval required between treatment

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[2] D–D is the trade name for a mixture of two isomers of 1–3 dichloropropene and 1–2 dichloropropane. The D–D used in these investigations was kindly furnished by the Shell Chemical Corporation of San Francisco, California.

and planting varied with the dosage and the plant used. D–D was found to be effective as a nematocide in a cold soil (30°–40° F.).

Tam (6) found D–D to be less effective in suppressing nitrification than was chloropicrin. Carter (2) found water solutions of D–D to be effective against the larvae of *Anomala orientalis* Waterh. and *Adoretus sinicus* Burm. in soil around the roots of actively growing nursery stock. Carter (3) reports that the 1–3 dichloropropene fraction of D–D is the most toxic but that there is synergism between this and other fractions.

FIELD EXPERIMENTS

1943

A comparison of D–D and chloropicrin as soil fumigants for vegetable crops was made at the Poamoho Experimental Farm, island of Oahu, in the fall of 1943. The study was a cooperative investigation between the University of Hawaii Agricultural Experiment Station and the Pineapple Research Institute. The comparison was made in a red lateritic soil in which a heavy population of the root-knot nematode had been built up by earlier crops of tomatoes, carrots, and beets. Four treatments, including the check, were randomized using 5 replications. Fumigants were injected September 1 and 2 at a depth of 5 to 6 inches with Vermorel injectors at staggered points 10.4 inches apart in rows 9 inches apart. The soil moisture content at depths of 2, 6, and 10 inches, respectively, was 27.0, 34.3, and 34.7 per cent at the time of application. The physical condition of the soil was poor and was not favorable for the most effective fumigation. No soil cover nor water seal was used.

The test crops used in the experiment were Morse's Bunching carrots and Victor tomatoes. Plantings were made September 10 on either side of an irrigation furrow which passed through the center of each plot. Carrot seed was planted on one side of the furrow and 20-day-old tomato seedlings transplanted on the opposite side. Fertilization and care were uniform, agreeing with the established practices at Poamoho.

TABLE 1.—*Comparison of the effect of soil fumigation with D–D and chloropicrin on the development of root knot and on the yield of carrots at Poamoho, Oahu, in 1943*

Treatment			Nematode infestation				Yield in pounds per plot	
			Percentage of plants in each class					
Material	Pounds per acre	No. of plants examined	None	Slight	Moderate to severe	Mean class value	Total	Marketable carrots
			0	1	2			
Check		685	0.0	30.9	69.1	1.72	5.3	3.8
D–D	200	948	23.3	59.9	16.8	0.91	14.4	12.6
D–D	400	944	80.0	17.0	2.9	0.22	17.7	16.1
Chloropicrin	400	1135	75.4	18.8	5.8	0.29	19.0	17.5
Difference required for significance at 5 per cent level						0.30	7.7	7.3

TABLE 2.—*Comparison of the effect of soil fumigation with D–D and chloropicrin on the development of root knot and on the yield of tomatoes at Poamoho, Oahu, in 1943*

Treatment		Nematode infestation						
Material	Pounds per acre	No. of plants examined	Percentage of plants in each class				Mean class value	Yield in pounds per plant
			None	Slight	Moderate	Severe		
			0	1	2	3		
Check	.....	93	0.0	1.1	32.2	66.7	2.68	1.00
D–D	200	95	22.1	64.2	12.6	1.1	0.89	2.11
D–D	400	69	55.1	39.1	1.5	4.3	0.61	2.23
Chloropicrin	400	102	51.0	42.2	3.9	2.9	0.56	1.94
Difference required for significance at 5 per cent level							0.41	0.67

Data on root-knot infestation of the tomatoes were obtained by removing alternate plants 26 days after transplanting. All green fruit was picked and weighed when the remaining plants were 11 weeks old. The carrots were harvested when 4 months old, examined for nematode infestation, graded, and weighed. Results are in tables 1 and 2.

Bacterial wilt, caused by *Bacterium solanacearum* E.F.S., destroyed 18.7 per cent of the tomato plants during the 11 weeks of the test. Fusarium wilt, caused by *Fusarium bulbigenum* var. *lycopersici* Woll. u. Reink., destroyed an additional 2.0 per cent of the plants. Data were obtained on the occurrence of these two diseases and are presented in table 3. A trend was observed for the incidence of Fusarium wilt to be less and that of bacterial wilt to be greater following fumigation with D–D. The incidence of both Fusarium wilt and bacterial wilt tended to be lower following fumigation with chloropicrin. The differences between treatments were not statistically significant, however.

The data from both the carrot and tomato tests show that D–D and chloropicrin materially reduced nematode injury. None of the treatments entirely eradicated nematodes, however. A few heavily infested plants were found in every plot. Comparable control of nematodes was obtained with D–D and chloropicrin at the rate of 400 lb. per acre. D–D at the rate of 200 lb. per acre markedly reduced nematode infestation but was not so effective as the 400 lb. rate in the carrot treatments.

Both the total and marketable yields of carrots were increased by the

TABLE 3.—*Influence of soil fumigation with D–D and chloropicrin on the occurrence of Fusarium wilt, Fusarium bulbigenum var. lycopersici, and bacterial wilt, Bacterium solanacearum, of tomatoes at Poamoho, Oahu, in 1943*

Treatment	Fusarium wilt	Bacterial wilt
	Per cent	*Per cent*
Check	4.3	18.0
D–D, 200 lb./acre	2.0	22.0
D–D, 400 lb./acre	0.6	29.2
Chloropicrin, 400 lb./acre	0.9	6.8

fumigation treatments. The lighter dosage of D–D produced striking gains which were not significantly exceeded by 400-lb. applications of either D–D or chloropicrin. The yields following application of 400 lb. of D–D and chloropicrin were similar. Yields of green tomato fruits were approximately doubled by the fumigants. There were no significant differences between the fumigated tomato plots.

1944

The 1944 experiments were conducted at the Poamoho Experimental Farm in an area similar to that used in 1943. A heavy infestation of nematodes had been built up in an earlier planting of tomatoes. Soil temperatures ranged from 65° F. to 80° F. during the course of the experiment. Lettuce was used as the test plant. Data were gathered on the rate, depth, and methods of D–D application.

Rate of application. The 1943 tests showed that D–D was as effective as chloropicrin in controlling the root-knot nematode, but more data were

TABLE 4.—*Influence of the rate of application of D–D on root-knot development and on the yield of lettuce at Poamoho, Oahu, in 1944*

Rate of application in pounds per acre	Nematode infestation: Percentage of plants in each class					Mean class value	Yield in pounds per plot
	None 0	Very slight 1	Slight 2	Moderate 3	Severe 4		
Check	19.3	35.4	16.0	13.8	15.5	1.53	2.96
200	59.3	39.1	1.6	0.0	0.0	0.41	7.28
300	68.9	24.4	2.8	2.8	1.2	0.36	7.32
400	71.9	27.4	0.4	0.4	0.0	0.29	7.54
500	79.6	20.4	0.0	0.0	0.0	0.19	8.22
Difference required for significance at 5 per cent level						0.58	2.54

needed on rate of application. The 1944 tests included treatments with 200, 300, 400, and 500 lb. D–D per acre. The fumigant was applied in a continuous band in open furrows and immediately covered with soil to a depth of 4 inches. The treatments were randomized and replicated 5 times. Seven days were allowed for fumigation and escape of the fumigant. Lettuce was then directly seeded in the furrows just above the point of application of the fumigant. The plants were harvested at the stage of maximum head development. Yields were determined and the plants classified for nematode infestation. The results are in table 4.

Nematodes were not completely eradicated by any of the treatments. The population was greatly reduced, however, as was indicated by the infestation ratings and by the increase in yield following treatment. The yield of lettuce was more than doubled with all rates of application. There were no significant differences between treatments.

Depth of application. The influence of depth of D–D application was determined at Poamoho, Oahu, using lettuce as the test plant. D–D was

TABLE 5.—*Influence of the depth of application of D–D on root-knot development, and on the yield of lettuce at Poamoho, Oahu, in 1944*

Depth of application	Nematode infestation					Yield in pounds per plot
	Percentage of plants in each class				Mean class value	
	None 0	Slight 1	Moderate 2	Severe 3		
Check	6.1	25.5	47.2	21.2	1.85	7.4
4 inch	59.3	39.1	1.6	0.0	0.43	14.4
6 inch	66.7	22.1	0.8	0.4	0.35	13.0
8 inch	64.0	34.7	1.0	0.0	0.37	15.4
Difference required for significance at 5 per cent level					0.31	5.2

applied with a Vermorel injector at the rate of 300 lb. per acre at 4-, 6-, and 8-inch depths. Lettuce was directly seeded one week following fumigation. The lettuce was harvested when the heads were at maximum development. Yields and severity of nematode infestation were determined. The results are in table 5.

As in earlier experiments the nematode infestation was greatly reduced through treatment with D–D. Differences in lettuce yields or nematode infestation were not found among the three depths of application.

Methods of application. Three methods of D–D application were compared at Poamoho, Oahu, in 1944. D–D was (1) injected with a Vermorel injector, (2) applied in a continuous band, and (3) applied in the irrigation water as an emulsion. The rate of application was 300 lb. per acre. Lettuce was grown as an indicator crop. The results are in table 6.

The injection and continuous band methods of application were equally effective in controlling nematodes as is shown by the reduction in infestation and by the increase in yield. Applying D–D as an emulsion in the irrigation water gave no better results than the check.

1945

Results with D–D have shown that its effectiveness as a soil fumigant is influenced by soil conditions at the time of application. The purpose of the

TABLE 6.—*Influence of the method of application of D–D on root-knot development and on the yield of lettuce at Poamoho in 1944*

Method of application	Nematode infestation						Yield in pounds per plot
	Percentage of plants in each class					Mean class value	
	None 0	Very slight 1	Slight 2	Moderate 3	Severe 4		
Check	7.7	44.4	21.8	16.2	1.0	1.73	4.1
Injection	76.2	22.9	0.4	0.4	0.0	0.25	10.8
Continuous band	41.3	41.7	10.0	4.3	2.6	0.83	9.8
In water	21.0	51.0	13.4	9.5	5.1	1.18	4.6
Difference required for significance at 5 per cent level						0.76	3.1

1945 tests was to determine the effectiveness of D–D in a porous soil with a high temperature. The test area selected was at Koko Head, Oahu, at an elevation of approximately 15 feet above sea level. The soil was porous, showing very little evidence of lateritic weathering, and had an extremely heavy nematode population. A tomato crop had just been removed, and undecomposed root galls were still present. The area was thoroughly plowed and disced before treatment. D–D was applied at the rates of 100, 200, and 400 lb. per acre. The treatments were replicated 4 times and were arranged in a Latin square. The D–D was applied in the plow furrow by means of a burette and was covered with the plow immediately following application. The moisture content as determined[3] from representative samples collected from approximately the 3-inch depth averaged 16.9 per cent at the time of D–D application. The total porosity averaged 49.6 per cent and the organic content 3.48 per cent at the time of application. The soil pH ranged from 5.8 to 6.3. The soil temperature was 86° F. at the 3-inch depth and 84° F. at the 6-inch depth.

Manoa lettuce was used as the test crop. Plantings were made 1 day, 3 days, and 7 days following fumigation to determine the persistence of the toxic effects. The plantings were randomized within the replications. The lettuce was directly seeded and was thinned to 9 inches between plants following emergence. Irrigation was by the furrow system but water was not permitted to flow from one treatment to another. The lettuce was harvested at the stage of maximum head development. Thirty plants were harvested from each plot together with the root systems. The roots were removed, classified for nematode infestation, and weighed. The lettuce heads from each plot were also weighed. The results are in tables 7 and 8.

TABLE 7.—*Effect of the lapse of time between application of D–D and planting on the yield, root weight, and nematode infestation of lettuce at Koko Head, Oahu, during the summer of 1945*

	Pounds of D–D per acre	Time between treatment and planting		
		One day	Three days	Seven days
Yield per plot, in pounds	Check	5.0	7.1	6.6
	100	5.2	7.6	5.2
	200	6.2	10.0	7.4
	400	11.3	11.9	10.9
Weight of roots, in grams	Check	133.0	137.8	129.5
	100	157.0	159.3	124.8
	200	150.8	180.8	137.8
	400	187.5	191.0	178.6
Nematode infestation index	Check	94.2	97.2	97.5
	100	90.7	98.3	99.7
	200	76.1	82.6	89.8
	400	57.5	66.9	67.5

[3] The soil moisture, porosity, organic content, and pH determinations were made by the Chemistry and Soils department of the University of Hawaii Agricultural Experiment Station.

TABLE 8.—*Influence of soil fumigation with D–D on the yield, weight of roots, and nematode infestation of lettuce grown at Koko Head, Oahu, in the summer of 1945*

Rate of application in pounds per acre	Yield in pounds per plot	Weight of roots in grams	Nematode index
Check	18.7	400	96.1
100	18.0	441	96.2
200	23.6	469	83.1
400	34.0	557	64.0
Difference required for significance at the 5 per cent level	1.0	88.0	4.0

Inhibitory effects were not observed even when lettuce was seeded the day following treatment (Table 7). The yield, root weight, and nematode infestation were similar for the 3 dates of seeding. The plants from the 3 dates of planting were combined from each replication when harvested and the resulting data analyzed statistically. The 100-lb. application did not control nematodes and resulted in no increase in yield. The 200-lb. application resulted in both a reduction in nematode infestation and an increase in yield. The yield following the 400-lb. application was approximately twice that of the check. Although the 200- and 400-lb. applications reduced the nematode infestation, the nematodes were not eradicated as is indicated by the high nematode indexes following these treatments.

SOIL COVERS AND THEIR EFFECT ON FUMIGATION

When applying a soil fumigant, the recommendation is frequently made that a soil cover be used to prevent the rapid escape of the fumigant. Several types of covers were tried in 1945 under uniform conditions in artificially infested beds. The beds were 10 feet long, 4 feet wide, and 18 inches deep. The walls were constructed of concrete tile, and the soil used was a mixture of 2 parts garden soil and 1 part coral sand. The pH of the soil was between 7.6 and 7.9, and the soil moisture at the time of fumigation was 14.3 per cent in the upper 2 inches and 16.1 per cent at the 6-inch depth. The beds were infested by thoroughly mixing 4 lb. of chopped, heavily galled tomato roots into the soil of each bed one week in advance of planting.

The D–D was applied at 1-foot intervals at a depth of 6 inches by means of a burette. Three and one-half cubic centimeters were applied per square foot which very closely approximated a 400-lb.-per-acre rate. Each bed was divided into 2 parts by means of a heavy board, thereby permitting 2 treatments to the bed. After the D–D had been applied the beds were given the following treatments: (1) Bed covered with glue coated paper. (2) Bed covered with 3 layers of newspaper and lightly wetted down twice a day. (3) Bed soaked with water twice a day. (4) Bed wet down to a depth of 2 to 3 inches. (5) No cover or water seal. (6) Check. No cover or D–D treatment.

The paper covers were removed and watering discontinued after the fifth day. Bounty tomato seed was planted in rows spaced 7 inches apart one

week following treatment. When the tomato seedlings were 5 weeks old they were removed and the roots examined for nematode gall formation. An examination of a minimum of 200 plants per treatment showed no nematode gall formation on the roots of plants from covered or watered beds. One small gall was observed on a single plant from the treatment receiving no cover or water seal. Plants removed from the nontreated check showed severe root galling and were less than half the size of the plants from fumigated beds.

EFFECT OF D–D ON THE INCIDENCE OF BACTERIAL WILT

An observation during the 1943 tests indicated that application of D–D might increase the incidence of bacterial wilt of tomatoes. To determine the validity of this observation the influence of D–D and chloropicrin on the incidence of the disease was measured under controlled conditions. A mixture of 2½ parts of garden soil and 1 part coral sand was placed in a tile-enclosed bed and heavily inoculated with a water suspension of *Bacterium solanacearum* grown in nutrient broth. Tomato plants grown in the soil were killed by bacterial wilt. Samples of soil were removed, placed in sealed drums, and fumigated with 7 different amounts of D–D and chloropicrin as listed in table 9. After allowing one week for fumigation, eight 1-gallon cans of soil were removed from each drum. Six tomato seedlings were transplanted to each can. The occurrence of bacterial wilt is recorded in table 9.

TABLE 9.—*Influence of D–D and chloropicrin on the incidence of bacterial wilt, Bacterium solanacearum, on tomatoes grown in the greenhouse in artificially inoculated soil at Honolulu, Oahu, in 1945*

Application in pounds per acre	Percentage incidence at 4 dates following transplanting[a]			
	19 days	27 days	38 days	55 days
Check	62.5	89.6	95.8	100.0
100 lb. D–D	31.3	66.7	81.3	91.7
200 lb. D–D	22.9	52.1	77.1	85.4
400 lb. D–D	6.3	18.8	31.3	91.7
600 lb. D–D	0.0	4.2	10.4	52.1
1000 lb. D–D	0.0	2.1	4.2	14.6
400 lb. chlor.	2.1	2.1	6.3	27.1
400 lb. chlor., 400 lb. D–D	0.0	0.0	2.1	20.8

[a] Determined from 48 plants grown in gallon cans (6 plants to the can).

All treatments reduced the occurrence of bacterial wilt as compared to the check. None of the treatments eradicated the organism, however. Chloropicrin at the 400-lb.-per-acre rate was more effective than D–D at the same rate. D–D at the 1000-lb. rate was the most effective of the treatments. With the 1000-lb. rate, an inhibitory effect on growth was noted for about 30 days following transplanting. The plants receiving a combination of D–D and chloropicrin were the most vigorous throughout the test.

DISCUSSION

In Hawaii the yield of vegetables has been increased with applications of D–D as low as 200 lb. per acre. The nematocidal properties of D–D are

comparable to those of chloropicrin, which is more costly and difficult to apply. Effectiveness of the fumigation is dependent on the soil type and condition, on environmental factors, and on the thoroughness of the application.

The results show that a definite recommendation regarding the rate of application cannot be made. A lower rate was required with the heavier soils at Poamoho than with the lighter, more porous soils at Koko Head. This difference in efficiency may also be partially attributed to rapid volatilization of D–D at the high soil temperatures which are prevalent at the lower elevations particularly during the summer months. The manufacturers have warned that effectiveness may be reduced at temperatures above 80° F.

Results from tests in tile-enclosed beds indicate that effective nematode control may be obtained without the use of a soil cover or seal following fumigation. Soil covers were not tried on soils with extremely high temperatures such as are found during the summer at the lower elevations. It might be expected that a good seal would retard the rapid escape of the fumigant under these conditions and thereby increase the efficiency of the fumigation.

The favorable yield response obtained with D–D is undoubtedly due to a combination of factors. Although the root-knot nematode infestation was reduced each time an improvement in yield was noted, the reduction was not always in proportion to yield increase. No study was made of biological responses other than nematode control and the preliminary study of Fusarium wilt and bacterial wilt control.

No data were secured on the persistence of treatment effects. In none of the field tests were all of the nematodes completely eradicated, however. Under the favorable Hawaiian environment the nematode population may be expected to build up rapidly when susceptible crops are grown. Other beneficial results may be more persistent in their effect.

SUMMARY

Yields of vegetables were increased and the root-knot nematode, *Heterodera marioni* (Cornu) Goodey, thoroughly controlled with D–D applications as low as 200 lb. per acre when conditions for fumigation were favorable. Under less favorable soil and environmental conditions the nematodes were only partially controlled with applications of 400 lb. per acre. The required rate of application was shown to be higher in a warm (84°–86° F.), porous soil than in a heavy, compact soil with a lower temperature. The continuous band and spot injection methods of application gave similar results. D–D applied as an emulsion in the irrigation water was ineffective. No differences were noted in the effectiveness of D–D with the shallow-rooted lettuce plant when the depth of application ranged between 4 and 8 inches. The nematocidal properties of D–D were found to be similar to those of chloropicrin.

The phytocidal effect of D–D was unimportant as was demonstrated by the absence of injury even though all the field plantings were made within a week following treatment. Beneficial effects from soil covers or seals were not demonstrated from tests made in artificially inoculated test beds.

The incidence of bacterial wilt, *Bacterium solanacearum*, was reduced in the greenhouse by soil fumigation with both D–D and chloropicrin. D–D was less effective than chloropicrin.

The root-knot nematode was not completely eradicated in any of the field treatments.

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A CORRELATION BETWEEN PATHOGENICITY AND CULTURAL CHARACTERISTICS IN THE GENUS ACTINOMYCES[1]

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INTRODUCTION AND REVIEW OF LITERATURE

From 1930 to date a number of graduate students in the Department of Plant Pathology at Cornell University have studied various phases of the Actinomyces-potato scab problem. From 1930 to 1936 the senior author collected and studied isolates of the genus *Actinomyces* in an attempt to establish their identity and, failing that, to devise a workable system of classification (19). From 1938 to 1941 the junior author, studying the same group of isolates, developed a method of obtaining reproducible pathogenicity trials and tested the pathogenicity of 127 of the isolates, as well as of a number of named species from type-culture collections (4). The present paper is a joint presentation of data pertinent to a correlation observed between the behavior of the isolates studied and their pathogenicity to potatoes in greenhouse trials.

Taxonomic studies within the group of soil and plant inhabiting Actinomycetes have been attempted by many investigators. Methods devised by Neukirch (13), Krainsky (10), Waksman and Curtis (22), Waksman (21), Millard and Burr (12), Jensen (9), Wingerberg (24), and Duché (5) have been found, with few exceptions, inadequate by subsequent workers. The senior author found himself able to identify positively only one of the 143 isolates by any system previously applied to the genus *Actinomyces*.

The causal relationship of *Actinomyces scabies* (Thax.) Güss, to scab of the Irish potato was demonstrated by Thaxter (20) in 1891. His findings on pathogenicity have been supported by subsequent workers, a few of whom have claimed certain other species of *Actinomyces* to be pathogenic to the potato. However, in practice, many workers have experienced difficulty in reisolating the organism used as inoculum and in maintaining freedom from scab in the noninoculated control plantings. The junior author attempted to devise a method whereby positive and reproducible tests of pathogenicity could be obtained and to test the pathogenicity of these isolates to determine whether any correlation existed between growth under controlled conditions and pathogenicity.

[1] In preparing this paper the authors have used the generic name of *Actinomyces* as accepted prior to the work of Waksman and Henrici (23). According to the latter system of classification, all of the isolates included in this study belong in the genus *Streptomyces*.

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The authors are indebted to Professors F. M. Blodgett and Walter H. Burkholder for advice and encouragement throughout these studies.

METHODS

Taxonomic work. One hundred and forty-three isolates of *Actinomyces* were used in the study reported, 100 of which were isolated by the senior author and 21 by other workers at the same laboratory. The remaining 22 isolates were obtained from other laboratories or indirectly through type-culture collections. Of the local isolates, 110 were obtained from scabbed potato tubers, 8 from scabbed beets, and 3 from scabbed radishes during the period 1930 to 1934. Taylor's method of isolation of *Actinomyces* from scabbed potato tubers (18) was used to obtain most of the local isolates.

Single colonies were fished from the isolation plates. The cultures so derived were plated repeatedly at high dilution, and very young colonies, after microscopic examination, were selected. That this method was satisfactory in maintaining purity of the isolates is shown by the fact that: (1) tests of a physiological nature were reproducible over a period of years; (2) culture 1, isolated in 1930, then used as inoculum on potato, and 1A3, the culture obtained upon reisolation from the resultant scab lesion, were found alike in all respects through 1936 and were similarly pathogenic in 1942; and (3) spore measurements from any culture were distributed according to a normal curve as would be expected from pure cultures (unpublished data compiled by Dr. F. M. Blodgett).

When transferring from stock cultures, which were grown on potato-dextrose agar, a loop of very fine platinum or Chromel-A wire was touched to the sporulating surface of the growth. The aerial spores thus obtained were used as transfer material, care being exercised to avoid transferring bits of the substrate to the culture solution.

Morphological studies were limited principally to tests for acid-fastness, Ziehl-Neelsen method, made by Dr. Ruth Gordon, then of the New York State Veterinary College.

The methods of study of physiological characteristics were those recommended in the Manual of Methods for Pure Culture Study of Bacteria issued by the Society of American Bacteriologists (16). All tests of the utilization of sources of carbon were conducted in solutions of neutral reaction. The slightly modified formula of Ayers, Rupp, and Johnson, as given in the Manual of Methods (17), was used with all of the carbon sources, with the exception of starch, phenol, and the organic acids. One per cent of the carbohydrate, alcohol, or glucoside was added to the basal medium. Tyrosine was used at 0.04 per cent. Arabinose, xylose, dextrose, levulose, galactose, sucrose, maltose, raffinose, and malonic acid were sterilized by passage through a Berkefeld N filter, added to the sterilized and cooled basal medium and thence siphoned aseptically to previously plugged and sterilized test tubes. The organic acids were prepared according to the method of Ayers, Rupp, and Johnson (1), modified according to Burkholder (2) in that 0.15 per cent of the sodium salt of the acid was used.

Growth on milk was tested according to the method of Clark and Lubs (3) using brom cresol purple as an indicator in fresh separated milk. In

order to detect pigment production in some groups, duplicate tubes were prepared without the indicator.

Agar-shake cultures were prepared with Bacto nutrient agar according to the manufacturer's directions. Following McCormack (11), no dextrose was added. Tubes of melted agar were stabbed to the bottom with the transfer material, agitated by rolling, plunged in ice water, and incubated at 27° to 28° C.

The utilization of paraffin was tested by adding a cube of approximately 0.02 grams of wax paraffin (melting point 52–54° C., J. T. Baker Chemical Company) to each tube of the non-carbon basal medium. The purified paraffin was prepared by Dr. D. K. O'Leary from similar paraffin, using the method of Piper *et al.* (14).

All carbohydrates, alcohols, and glucosides used were of the highest purity obtainable from the Pfanstiehl Company or from the Difco Company. The organic acids were of the C.P. grade furnished by the Pfanstiehl Company or by the Baker Company.

Starch hydrolysis was tested according to the recommended method and also by the addition of 0.04 per cent of soluble starch to the semi-solid medium of Zobell (25). Tests of nitrate reduction also were made by the recommended method. The method of Zobell (25), who added zinc dust to the culture tube to check negative nitrite tests, also was used.

Maximum temperature permitting growth was determined by the incubation of streaked slants of potato-dextrose agar for seven days at 32°, 35°, 40°, 42°, and 46° C. No provision was made to avoid drying of the medium.

Pathogenicity trials. The experimental procedures employed in the pathogenicity studies were those of Goss (8), with several major adaptations.

The experiments were conducted in a greenhouse held at temperatures ranging from 20° to 30° C. Before each experiment the room (walls, ceiling, benches, and floor) was washed with water to remove all soil and dust and then washed with a formaldehyde solution. During the course of each experiment the floor was kept damp by frequent sprinkling with water to avoid dust.

Scab-free tubers of the Green Mountain or of the Irish Cobbler variety were cut to 40 gram seed-pieces, treated with formaldehyde, covered one hour, and dried. When dry, the seed pieces were placed in a dark moist chamber at room temperature to permit suberizing and sprouting.

The soil, a mixture of two-thirds fertile loam and one-third peat, was placed in 6-inch clay pots, each with a 6-inch saucer, and sterilized for 3 hr. at 20 lb. pressure on two successive days. The sterilized pots of soil were allowed to cool in the sterilizer and the plantings were made as the pots were removed.

The inoculum was prepared by placing on the surface of potato-dextrose agar in Petri dishes one ml. of water suspension of the organism taken from an agar slant. The Petri dishes were incubated at room temperature (20° C.) for 10 to 21 days, depending upon the time required for the culture to

cover the surface of the plate. The inoculum from two Petri dishes was used in each 6-inch pot.

As the pots of soil were removed from the sterilizer, the upper portion of the soil was removed. The sprouted seed-piece was placed in the center of the pot about 3 inches from the top, and the inoculum was washed from the Petri dishes to the seed-piece and the surrounding soil. The removed soil was replaced to within one inch of the top of the pot. Four pots of each isolate were planted in most cases. After the planting of each isolate the hands and tools were washed in 70 per cent alcohol and the tools flamed. Control plantings were made in the same manner with the omission of the inoculum. The pots were then placed at intervals of 6 inches on the greenhouse bench. All watering was done by adding water to the saucer rather than directly to the soil. This permitted the maintenance of a dry soil surface which would be unfavorable to the growth of spores or fragments of mycelium which might be found in the air.

At maturity the tubers were harvested and the number of scabbed tubers was recorded.

RESULTS

Taxonomic work. Of the isolates tested, 128 were non-acid-fast. Fifteen were partially acid-fast in that acid-fast elements were present in the preparations but were accompanied by non-acid-fast elements (7).

Starch was hydrolyzed by all of the 128 non-acid-fast isolates and by none of the 15 acid-fast isolates.

The production, within a few days, of a dark brown surface ring of growth on milk, with or without an indicator, characterized 66 isolates, all non-acid-fast. The color of this ring lies within the following limits in Ridgway's (15) system of color classification: 15′ i–m, to 17′ i–m. The intense brown pigment produced by the members of this group obscured the color of the indicator and, in many cases, necessitated electrometric determination of the reaction which varied considerably within the group. Of the remaining 77 isolates, 71 produced a distinctly alkaline reaction; four produced a hard, acid curd approximately 14 days after transfer, and it endured for at least three months; one produced no visible change in reaction in the presence of abundant growth; and one did not grow on milk.

On agar-shake tubes microaerophilic development of colonies occurred with 13 of the acid-fast isolates. With these isolates a dense ring of colonies developed at from 5 to 10 mm. beneath the surface of the agar. As the cultures aged, additional rings developed beneath the original ring. This microaerophilic growth always was accompanied by growth on the surface of the agar. With the non-microaerophilic isolates, no growth developed below 5 mm. from the surface, and such subsurface development was continuous with the surface growth. In most cases the subsurface growth was very shallow.

Gelatine was liquefied by all of the non-acid-fast isolates and by the two

acid-fast isolates which were not microaerophilic. The 13 microaerophilic acid-fast isolates did not liquefy gelatine.

In the "dark brown ring on milk" group, the distinction between positive and negative tests of nitrate reduction was very definite on Zobell's medium. Among the other 77 isolates, many yielded clear-cut tests; but some isolates produced a very weak reaction which tended to be variable between replicates and between dates of testing.

The utilization of sources of carbon proved to be a reproducible characteristic of the isolates tested. The ability to utilize sources of carbon varied with the different isolates so that separatory distinctions could be made. Definite growth on carbohydrates, alcohols, glucosides, and paraffin was accompanied by the production of an acid reaction in the culture solution; with the organic acids an alkaline reaction was produced. A reaction change of greater than 0.5 pH was associated with acceptable growth.

Succinic, formic, oxalic, and citric acids were more useful than any other of the 30 carbon sources tested in separating the isolates. Several sources were utilized by the majority of the isolates, while salicylic acid was utilized by a single isolate. Malic acid was the only source utilized by all isolates, while phenol at 0.10 per cent was utilized by no isolate.

All isolates produced ammonia and no isolate produced indol under the conditions tested.

The temperature tolerance of any isolate remained constant in these studies. All isolates grew well at 32° C., 9 failed to grow at 35° C., 101 grew at 35° C. but failed to grow at 40° C., while 25 grew at 40° C. but not at 42° C. Eight isolates grew at 42° C., two of which grew at 46° C. but appeared at that point to be near the upper limits of temperature permitting their growth. It was characteristic of all isolates that growth was abundant at very close to the upper limits of their heat tolerance.

One of the local isolates, No. 7, was similar in all respects to isolate No. 3502, obtained from the type-culture collection at Baarn as *Actinomyces tricolor* of Wollenweber. These isolates both appeared to be identical with the earlier-described *Actinomyces violaceus-ruber* of Waksman.[3] These isolates are characterized by the production of anthocyanin (6).

The accompanying taxonomic key to the isolates studied was prepared on the basis of the tests herein discussed. Pending determination of the physiological characteristics of the many named species available in type-culture collections, it was deemed inadvisable to apply names to any of the isolates separated in this key.

Pathogenicity trials. Lesions of the common scab of potato vary in their final form from raised lesions to shallow to deep pits; all lesions are shallow in their early stages. Typically they are circular, with a tendency to develop concentric rings of cork as they become older. Their margin is definite. In addition to typical lesions of the common scab of potato,

[3] The statement as to the similarity of *Actinomyces tricolor* to *Actinomyces violaceus-ruber* is based on correspondence with Dr. Selman A. Waksman, who had compared the two isolates, and also on comparisons made at the authors' laboratory.

KEY TO THE ISOLATES STUDIED

ISOLATE NO.

A not acid-fast, starch hydrolyzed
B dark brown surface ring on milk
C nitrite not present on Zobell's medium
D formic +
E tartaric – 27b
EE tartaric +
F milk acid 12aB, 409
FF milk alkaline 63b
DD formic –
E milk alkaline
F oxalic +
G tartaric + ... 3500
GG tartaric – C14
FF oxalic –
G rhamnose + ... 79, 80a
GG rhamnose – 27ay
EE milk acid
F succinic – 53b
FF succinic +
G nitrite present syn. NO_3 solution 1, 1A3
GG nitrite not present syn. NO_3 solution
H oxalic + ... 48aW
HH oxalic – 28a
CC nitrite present on Zobell's medium 15, 16, 9b, 17, 27d, C7, C48, 61b, 47esm, Cs28, 46, 74, 75, 76, 412, 84a, 85a, 24, S2, 19, 45, 9a, N54, 29, 53a, Cs8, 42, 18sm, N23, 28b, 39, B11, C6, 41, 14, 25, 27ax, 36, 54a, 47ew, C28, 18w, 62, 64a, 64b, 65b, 68b, 73, 407, 410, 78, 81a

A not acid-fast, starch hydrolyzed
BB no dark brown ring on milk
C succinic +
D formic +
E oxalic + ... 3502, 7
EE oxalic –
F sucrose + ... 59
FF sucrose –
G malonic + ... 88
GG malonic –[a]
H paraffin +[b] 49b, 6B, 9e, 3307, 6W, C18pw, C11, 67, 427
HH paraffin – 3504
DD formic –
E oxalic +
F citric +
G milk acid, curd, scarlet ring 80b
GG milk alkaline
H sucrose +
I raffinose + ... C5, 13b
II raffinose –
J max. temp. 40°, salicin – 63a, 63d
JJ max. temp. 35°, salicin + ... 57b
HH sucrose –
I tartaric + ... 3369B
II tartaric –
J malonic, arabinose, lactose + ... 47a
JJ malonic, arabinose, lactose – 82
FF citric –
G nitrite, Zobell med. –, salicylic + ... 81b
GG nitrite, Zobell med. +, salicylic – C13
EE oxalic –
F sucrose +
G milk acid, curd 49a
GG milk alkaline
H nitrite, Zobell medium + ... 50
HH nitrite, Zobell medium – 56

FF sucrose –
G max. temp. 35°
H tyrosine agar red 47b
HH tyrosine agar uncolored 54b, 48ab, 3005, 2745, 55a, 54c, 2744, 66, 8
GG max. temp. 40° or above
H milk acid, curd 31, 13a
HH milk alkaline
I citric – 55b
II citric +
J sucrose + ... 3329
JJ sucrose –
K rhamnose – 68a
KK rhamnose +
L paraffin + ... 3006
LL paraffin – 1356, 423, 426

A not acid-fast, starch hydrolyzed
BB no dark brown ring on milk
CC succinic –
D oxalic + ... 84b
E citric –
EE citric +
F max. temp. 35°
G milk alkaline 89
GG milk, no growth 91
FF max. temp. 40° 57a
DD oxalic –
E citric –
F rhamnose + ... 90
FF rhamnose – 87
EE citric +
F rhamnose –
G dextrose + ... 3352, 3503
GG dextrose – N4, N3
FF rhamnose +
G paraffin +
H growth above 42° 63c
HH max. temp. 35° 3501
GG paraffin –
H nitrite, syn. NO_3 med. – 65a
HH nitrite, syn. NO_3 med. +
I sucrose + ... 69
II sucrose – 3372

AA acid-fast (partially), starch not hydrolyzed
B gelatine not liquefied, microaerophilic
C reaction of milk unchanged 10
CC milk alkaline
D max. temp. 40°, paraffin + ... 34, 20, 21a, 27c, 48c, 61a, 72
DD max. temp. 34°, paraffin – 22, 30, 52, 55d, 83
BB gelatine liquefied, not microaerophilic
C milk cleared, no acid on purified paraffin 71
CC milk not cleared, acid on purified paraffin 86

[a] No growth, or growth without change in reaction.
[b] Throughout this key paraffin, unless otherwise specified, refers to the unpurified form.

various types of russeting frequently are encountered on potato tubers. These russeted areas are superficial and usually are irregular, with shallow cracks not in a concentric pattern. Rarely do these areas have a definite margin. Repeated attempts to isolate a causal organism from beneath the periderm covering the russeted areas of tubers have not been successful.

Typical potato scab, as distinguished from superficial russeting of the surface, was produced by all of the 61 "dark brown ring" isolates tested. No member of the 66 "non-dark brown ring" isolates tested caused typical

scab on potato tubers in these experiments. Throughout the course of these experiments no scab lesions developed on the 684 tubers grown in noninoculated control pots.

Within the "dark brown ring" group, variations occurred in the amount of scab produced on the tubers, though all produced some scab infection. Of the total of 1178 tubers produced following inoculation with these 61 isolates, 763 tubers or 64.8 per cent of the total were scabbed.

The isolates studied did not differ consistently in type of scab lesion produced. In many cases scab lesions varying from shallow lesions to deep pits were found among the replicated pots of one isolate, and frequently within the same pot or on the same tuber. It is emphasized that these tubers were grown under conditions continuously favorable for the inoculum and that the varieties of potato used were highly susceptible. More resistant varieties under conditions less favorable for the inoculum would be more likely to show relationships between the species or strain of *Actinomyces* and specific varietal response.

Superficial russeting, not considered typical of potato scab, developed on 52 of the 258 tubers produced in the trials of the acid-fast isolates, and on 40 of the 1050 tubers in the non-acid-fast, non-brown ring isolates. It was more abundant with certain isolates in the latter group; 12 of the 40 tubers occurring among 72 tubers from tests of one isolate. By the method of isolation used (18) it was possible to reisolate from the russeted areas the organism used as inoculum in the acid-fast group but not in the non-acid-fast group. This russeting was not observed in the trials of the "dark brown ring" isolates.

DISCUSSION

The correlations observed between pathogenicity and the behavior on milk held over a wide selection of Actinomycetes throughout these studies. If true of all members of the genus *Actinomyces,* it offers a simple laboratory method of testing probable pathogenicity. Only further trials can determine the validity of this finding.

A superficial russeting of the surface of the tubers was associated with inoculation by a number of isolates. This russeting was distinct from the reaction usually considered as typical of potato scab. Furthermore, it was associated with isolates of widely different types, none of which induced the typical scab reaction. Reisolation of the acid-fast isolates from these russeted areas offers no proof of pathogenicity since the isolation method employed was based on methods for the isolation of the acid-fast Mycobacteria. It is possible that this technique might result in reisolating the acid-fast inoculum from the surface of the tuber, rather than from within the tissues. On the basis of these studies the cause of this russeting cannot be considered as established.

SUMMARY AND CONCLUSIONS

Studies were made of the physiological characteristics of 143 isolates of

the genus *Actinomyces*, mostly from scabbed potato tubers, and their pathogenicity to potato tubers.

A satisfactory method of testing the pathogenicity of Actinomyces isolates on potatoes was developed.

Within the group studied, the ability to produce typical lesions of potato scab was correlated perfectly with the production of a dark brown ring of surface growth on separated milk.

Partial acid-fastness was correlated perfectly with non-microaerophilic growth.

Separatory differences were found in the response of the 143 isolates on 28 of 30 different sources of carbon and in a number of other tests of bacteriological nature.

A russeting of the surface of the tubers was associated with inoculation by certain otherwise non-pathogenic isolates. The causal nature of this association is not considered as having been proven.

A key to the isolates studied, based upon tests of a bacteriological nature, is presented.

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RESISTANCE TO CROWN RUST IN FESTUCA ELATIOR AND F. ELATIOR VAR. ARUNDINACEA[1]

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INTRODUCTION

Crown rust, *Puccinia coronata* Cda., is one of the most destructive diseases of *Festuca elatior* L. in the northeastern United States. Although meadow fescue is a minor component of many hay and pasture mixtures, it can be grown almost anywhere in this region (5), consequently, it is found abundantly in meadows and along roadsides. General distribution of the host, therefore, offers ample opportunity for widespread attacks of the disease. Fortunately, the variety of crown rust that attacks *F. elatior* is restricted to several species of grasses and is seldom likely to attack oats (1, 3).[3] According to Brown (1) *Puccinia coronata* may be sub-divided into seven varieties depending upon grasses attacked. She concluded, as had several earlier workers, that var. *lolii* and var. *festucae* are difficult to differentiate and are probably physiologic races of the same variety of rust. In the present experiments no effort was made to differentiate the varietal or physiologic forms of *P. coronata*.

In recent years a coarse-leaved, large growing fescue known as "tall" fescue [*F. elatior* var. *arundinacea* (Schreb.) Wimm.] has been utilized as a forage grass. Tall fescue, although not tested so extensively as meadow fescue, has been observed by Piper (5) and Hardison (2) to be highly resistant to crown rust. As a result of preliminary observations and greenhouse tests a program of selection and breeding was initiated to secure resistance to crown rust in plants of *F. elatior*.

MATERIALS AND METHODS

Single tillers were usually established from plants moved from the field to the greenhouse for further testing. Most of the clones were tested in triplicate to reduce variation in infection to a minimum. Plants were also established from seeds of different collections by sowing them in flats of steamed soil and later transplanting individual seedlings to soil in 3-inch pots.

Urediospores were obtained in several ways. Leaves with mature uredia were collected in the field and stored in a refrigerator. Urediospores were also collected from leaves of heavily rusted plants by gently shaking the plants over a large evaporating dish. The spores were transferred to vials

[1] Contribution No. 80 of the U. S. Regional Pasture Research Laboratory, Division of Forage Crops and Diseases, Bureau of Plant Industry, Soils, and Agricultural Engineering, Agricultural Research Administration, U. S. Dept. of Agriculture, State College, Pa., in cooperation with the northeastern states.

[2] Associate Pathologist and Geneticist, respectively.

[3] A sample of crown rust used in this investigation was sent to Doctor H. C. Murphy who found the urediospores noninfectious for differential varieties of oats.

by means of a camel's hair brush and stored in a refrigerator until used. Heavily rusted plants provided another source of inoculum since they were easily transplanted from the field to pots in the greenhouse. Abundant uredial infection was maintained on leaves of susceptible plants by periodically rubbing diseased and healthy leaves and then incubating the plants in a moist chamber.

For inoculation purposes, plants were atomized with a composited suspension of urediospores in tap water, and the inoculated plants were then incubated 48 hr. in a moist chamber. After removal from the moist chamber the plants were placed on a bench in the greenhouse for development of uredia. Severity of rust and infection type were read 10 to 15 days following inoculation. Murphy's classification of infection types for crown rust of oats (3) was adopted with slight modifications. All very susceptible plants were placed in class 3. This included all plants having abundant, moderately large to large uredia with or without necrosis or chlorosis immediately surrounding the uredia. Most susceptible plants observed developed chlorotic areas immediately surrounding the uredia. Plants that reacted doubtfully to inoculation were clipped and reinoculated after new noninfected leaves developed.

RESULTS

During the summers of 1942 and 1943, spaced plants of *Festuca elatior* and *F. elatior* var. *arundinacea* were observed for resistence to crown rust. Of several thousand plants of *F. elatior* examined in the nursery only a few showed evidence of resistance. The resistant clones were transplanted to the greenhouse and tested further during the winter of 1943 and 1944. Artificial inoculation confirmed field observations that clones of *F. elatior* in general are susceptible to crown rust while clones of *F. elatior* var. *arundinacea* are usually resistant. However, several plants of *F. elatior* observed to be free of rust in the field proved immune when tested in the greenhouse. Seeds which produced the rust-immune plants (247–1 and 247–2) had been collected by Doctor F. H. Steinmetz, Department of Botany, University of Maine, in a meadow near Orono, Maine. In figure 1, healthy leaves from a rust-immune plant of *F. elatior* are compared with rusted leaves from a susceptible plant.

Additional seeds were collected by Doctor Steinmetz from plants growing in this meadow and adjacent fields during the summer of 1944. Plants obtained from these seeds as well as plants secured from several other seed collections were tested for susceptibility to crown rust during the following winter.

Cytological studies are being reported in detail in another paper, by Myers and Hill (4). Hence only the data on chromosome numbers will be presented here (Table 1).

Data presented in table 1 came from testing approximately 20 plants of each strain of fescue derived from single seeds of a collection. In most

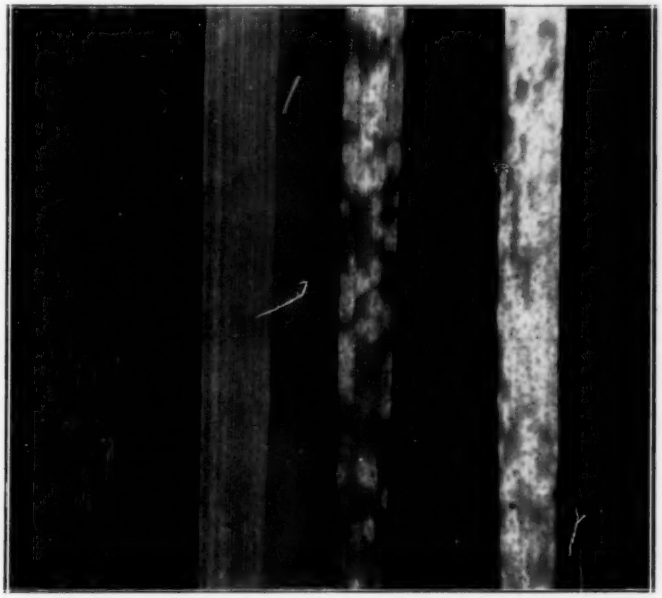

FIG. 1. Leaves from a rust-immune plant of *Festuca elatior* on the left and a rust-susceptible plant on the right.

cases, the 14-chromosome *F. elatior* was susceptible to crown rust while the 42-chromosome *F. elatior* var. *arundinacea* was resistant. Both the Svälof and Otofte strains yielded plants of variable rust reaction. Some plants appeared resistant to rust while others were extremely susceptible. However, none of the resistant plants had the immunity to crown rust characteristic of the Maine rust-immune plants. Variability within the Svälof and Otofte strains suggests that crown rust-resistant individuals might be selected from this material.

When plants obtained from seeds of fescue harvested in the vicinity of the original Maine rust-immune plants were tested, no further 14-chromosome, immune plants were discovered. A summary of the rust reactions of

TABLE 1.—*Crown rust reaction and chromosome number of some collections of Festuca elatior and F. elatior var. arundinacea*

Collection	Reaction to crown rust	2*n* chromosome number
Festuca elatior		
Maine 247–1 and 247–2	Immune	14
Svälof early strain	Resist. to susc.	14
Otofte strain	Resist. to susc.	14
Minn. 1449	Susceptible	14
S–53 (Wales)	Susceptible	14
F. elatior var. *arundinacea*		
N.Y. 2659 (England)	Immune to highly resistant	42
Alta (Ore. F.C. 29,366)	Immune to highly resistant	42
Suiter strain (Ky. K–31)	Immune to highly resistant	42

these plants is in table 2. One collection, (44–6), consisted of 42-chromosome plants which were preponderantly immune from crown rust; the other collections were 14-chromosome plants which were mostly susceptible.

TABLE 2.—*Crown rust reaction and chromosome number of three fescue collections from Maine*

Collection	Number of plants in each rust reaction class[a]					Total	$2n$ chromosome number
	I	0	1	2	3		
Maine 44–6 (27 heads collected from meadow near original pasture)	1130	31	135	5	12	1313	42
Maine 44–7 (10 heads collected from old lawn adjacent to original pasture)	0	1	0	5	282	288	14
Maine 44–8 (8 heads collected from original pasture)	0	0	1	25	214	240	14

[a] I—Immune—no macroscopic evidence of infection.
0—Nearly immune—no uredia formed; necrotic areas or chlorotic flecks present.
1—Highly resistant—no uredia or uredia few, small, always in necrotic areas; necrotic areas often produced without development of uredia.
2—Moderately susceptible—uredia fairly abundant, small to midsized, always in necrotic or chlorotic areas.
3—Susceptible—uredia abundant, midsized to large with or without necrosis or chlorosis immediately surrounding the uredia.

DISCUSSION

It has been shown experimentally that 14-chromosome *Festuca elatior* is usually susceptible to crown rust, *Puccinia coronata*, while 42-chromosome *F. elatior* var. *arundinacea* is usually resistant. That some rust-resistant germ plasm exists in *F. elatior* is demonstrated by the variability in rust reaction encountered in the varieties Svälof and Otofte. Since only one collection of rust-immune *F. elatior* was encountered among all the material tested the natural occurrence of rust-immune *F. elatior* appears to be rare. Additional collections secured from the same and neighboring fields proved to be either *F. elatior* in which most of the plants were susceptible to crown rust or *F. elatior* var. *arundinacea* in which most plants were resistant. The 14-chromosome condition of the rust-immune material should aid in transferring rust resistance to strains of *F. elatior* that are agronomically desirable but susceptible to *Puccinia coronata*.

SUMMARY

Most collections of meadow fescue, *Festuca elatior*, are susceptible to crown rust, *Puccinia coronata*, while those of tall fescue, *F. elatior* var. *arundinacea*, are usually resistant.

One collection of meadow fescue obtained from Maine was immune from crown rust.

Additional material from the same and neighboring fields proved to be either meadow fescue in which the majority of plants were susceptible to

crown rust or tall fescue which varied in rust reaction from susceptible to immune.

U. S. REGIONAL PASTURE RESEARCH LABORATORY,
STATE COLLEGE, PENNSYLVANIA.

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5. PIPER, C. V. Forage plants and their culture. Macmillan Co., New York. 1924.
6. SEMPLE, A. F., H. N. VINAL, C. R. ENLOW, and F. E. WOODWARD. A pasture handbook. U. S. Dept. Agr. Misc. Publ. 194. 1934. Rev. 1942.

PHYTOPATHOLOGICAL NOTES

Pinto Leaf, a Transmissible Disease of Cherry.—During a survey of cherry orchards at The Dalles, Oregon, in June, 1943, two Napoleon (Royal Ann) trees were discovered with unusual chlorotic leaves. Since the mosaic symptoms on the diseased leaves resemble the blotchy pattern of certain western horses known as pinto ponies, the name "pinto leaf" has been selected as a descriptive common name. In subsequent surveys at The Dalles the disease also was found on the varieties Napoleon, Black Republican, and Stark Gold, as well as on mazzard seedlings, and later two infected mazzard seedling trees were discovered at Hood River, Oregon. Pinto leaf is of minor economic importance at the present time because of its limited occurrence.

Large to small chlorotic patches form on diseased leaves. The original pale green to yellow color of diseased tissue gradually changes to bright yellow or white. Any part of the leaf may be affected, but the chlorosis rarely forms a specific pattern (Fig. 1, A). Leaf symptoms are sometimes obscure or meager, especially on mazzard seedlings, in which the chlorosis often appears as a coarse stippling. Leaves of terminal shoots rarely become chlorotic, but when this occurs a few of the basal leaves may show signs of infection late in the season. Severely diseased trees appear slightly dwarfed, produce less new growth, and the foliage appears somewhat ruffled when viewed from a distance.

The fruit of Napoleon and Stark Gold on affected trees never attained proper maturity. When healthy Napoleon fruits were fully colored and sweet, diseased fruits remained yellowish-green and insipid in taste. The fruit from affected Napoleon trees had rarely been picked, according to the owner, because of its lack of color and inferior quality. Infected Stark Gold yielded smaller fruit of inferior flavor. The color differences have not been noticeable in any infected black cherry variety, but trees with symptoms as severe as in the Napoleon variety have not yet been observed.

Buds from diseased pinto-leaf trees were inserted into 13 symptomless sweet-cherry seedlings late in June, 1943. Definite symptoms of pinto leaf appeared in 3 of the seedling trees during the spring of 1944. In a second attempt at transmission during August, 1944, a series of buds taken from symptomless terminal shoots of diseased trees was inserted at Hood River into 20 healthy mazzard seedlings. A second group of 24 seedling trees was budded with infected heel spurs that had produced visibly infected leaves. Only one seedling of the first group became infected although several of the original buds formed new growth with clean foliage. Of the "spur bud" group, 5 seedlings developed typical symptoms, indicating that the virus probably is more concentrated in older growth or that it moves into terminal shoots rather tardily. Transmission to the seedling trees was apparent early in the spring of 1945.

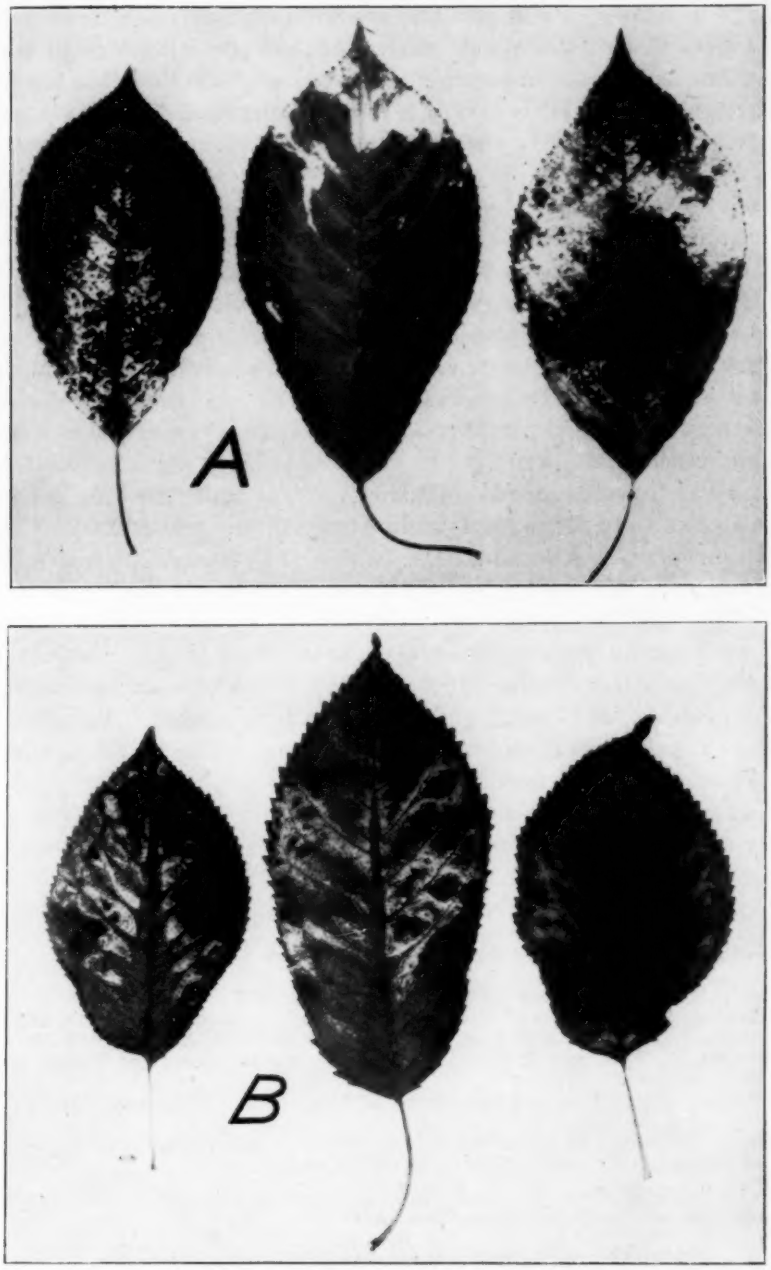

FIG. 1. Pinto leaf virus symptoms compared with "white spot" on cherries. A. Pinto leaf on Napoleon cherry leaves. B. White spot, a nontransmissible malady, on Black Republican cherry leaves.

The pinto-leaf symptoms on cherry resemble apple mosaic,[1] grape mosaic,[2] peach calico,[3] and a "white spot" disease of cherry,[4,5] but are distinct from line pattern as reported on cherry.[6] The characteristic "oak leaf" pattern of white spot (Fig. 1, B) generally serves to distinguish it from pinto leaf. Buds from typical white spot found on a Black Republican tree at The Dalles failed to transmit or perpetuate the symptoms when worked onto mazzard seedlings. The results substantiate those of Thomas and Rawlins[5] and Cochran[7] that white spot is not an infectious disease.

Attempts were made to transmit pinto leaf to sour cherry, peach, plum, apricot, chokecherry, rose, apple, and pear. A few fugitive leaf symptoms were observed only on one peach seedling, but they resembled neither those of cherry pinto leaf nor of peach calico. Under dry land farming conditions existing at The Dalles, successful bud unions form with difficulty, and this may account, in part, for the relatively small number of successful transmissions in that district.

Pinto leaf symptoms are distinct from any known disease affecting sweet cherries. For those desiring a Latin binomial, the name *Marmor pintofolium* is proposed.—J. R. KIENHOLZ, U. S. Fruit Disease Laboratory, Hood River, Oregon.

Virus Transmission by Cuscuta sandwichiana.[1]—Since Bennett[2] published the first paper on the subject in 1940, *Cuscuta subinclusa* Dur. and Hilg.,[2] *C. californica* Choisy,[2] and *C. campestris* Yuncker,[3,4] have been recorded as capable of transmitting plant viruses. The fourth species, *C. sandwichiana* Choisy, is reported here.

Cuscuta sandwichiana is an endemic species in Hawaii occurring occasionally along the coastal dunes or swamps. This dodder is not so aggressive a parasite as some of the other species and could not establish itself

[1] Bradford, F. C., and Lloyd Joley. Infectious variegation in the apple. Jour. Agr. Res. [U.S.] 46: 901–908. 1933.

[2] Hewitt, W. B. A graft-transmissible mosaic disease of grapevine. Phytopath. 35: 940–942. 1945.

[3] Blodgett, Earle C. Peach calico. Phytopath. 34: 650–657. 1944.

[4] Rhoades, A. S. Virus and virus-like diseases of sweet cherry in Utah, and notes on some conditions affecting various fruit crops. Plant Dis. Reptr. 29: 6–19. 1945.

[5] Thomas, H. Earl, and T. E. Rawlins. Some mosaic diseases of Prunus species. Hilgardia 12: 623–644. 1939.

[6] Willison, R. S. A line-pattern virosis of Shiro plum. Phytopath. 35: 991–1001. 1945.

[7] Dr. L. C. Cochran of this Division informed the writer he has made numerous attempts to transmit white spot in California without success.

[1] Published with the approval of the Director as Technical Paper No. 170 of the Pineapple Research Institute, University of Hawaii.

[2] Bennett, C. W. Acquisition and transmission of viruses by dodder (*Cuscuta subinclusa*). (Abstr.) Phytopath. 30: 2. 1940.

[3] Johnson, F. Transmission of viruses by the parasitic activities of dodder. (Abstr.) Phytopath. 31: 13. 1941.

[4] *C. repens* has also been reported by E. M. Hildebrand (The dodder vector of woody plant viruses. U. S. Dept. Agr., Pl. Dis. Reptr. 29: 196–197. 1945). The writer was informed by N. J. Giddings, who supplied the seeds to Hildebrand, that there had been some confusion in labeling and *C. repens* reported by Hildebrand should be known under the name *C. campestris*.

under greenhouse conditions, on several plants tested. These plants were *Dendrobium sp., Commelina diffusa* Burm. f., *Emilia sonchifolia* (L.) DC., *Nicotiana glutinosa* L., and pineapple. It established with difficulty on mature stems of tomato but never on young plants. It thrived, however, on cucumber, *Boerhaavia diffusa* L., and *Asystasia gangetica* (L.) T. Anders.; on the last plant a stock colony of the dodder was maintained.

The cucumber mosaic virus was very easily transmitted from cucumber to cucumber when dodder growing on the infected plants was attached to healthy plants. Twenty-one test plants were successfully infected. None of 19 other test plants became infected when the terminal cuttings from dodder grown on the infected plants were attached to them. It appears that the Hawaiian dodder is not so efficient as other dodders in retaining the virus. A mosaic, of which the identity has not been established, was commonly found on the island of Oahu on *Boerhaavia diffusa* which is an indigenous littoral herb. The virus was not transmitted into 29 test plants through the dodder connection.

Repeated trials were all failures in transmitting the spotted wilt virus from tomato to tomato. The dodder never established on young tomato plants with fresh symptoms; however, it did on mature plants with old symptoms. Forty-two mature test plants thus connected with such source plants did not demonstrate successful virus transmission. Since the spotted-wilt virus has been known to be difficult to acquire from the plants with old symptoms through either sap or insect transmission, the present data are not conclusive. The spotted-wilt virus, however, appears to be comparatively difficult to transmit through dodder vectors, as Bennett[5] reported this to be the case with *Cuscuta subinclusa, C. californica,* and *C. campestris,* particularly through the first two species.

Another indigenous parasitic vine, *Cassytha filiformis* L., which is much coarser than *Cuscuta,* has been proved to be unsatisfactory for transmission work on herbaceous plants.—K. SAKIMURA, Pineapple Research Institute, Honolulu, Hawaii.

Sclerotium rolfsii Sacc. and its Perfect Stage on Climbing Fig.—Climbing fig (*Ficus pumila* L.) is commonly used in Florida to cover stone or brick walls because it grows rapidly, is evergreen and is seldom attacked by insect pests or fungus diseases. During July, 1945, *Sclerotium rolfsii* Sacc. was found on several areas of dead leaves, noted on the north wall of the Plant Pathology greenhouse at Gainesville. These areas were roughly semi-circular with a radius of 6 to 18 inches and with the flat side of the semicircle at the ground. Mycelium was especially conspicuous on the tender, green stems and the underside of the leaves. White to pale mycelium covered the affected host organs, fastening adjacent leaves together in some cases and spreading out fan-like over newly affected leaf areas. Small,

[5] Bennett, C. W. Studies of dodder transmission of plant viruses. Phytopath. 34: 905–932. 1944.

nearly spherical sclerotia were formed on the dead, young stems and along the edges of affected leaves. Most of the dead leaves were reddish-brown, although those most recently affected were brownish-olive. Young stems and leaves were killed outright. Older stems, from which the leaves had fallen, were not killed, and they produced new sprouts later in the season. Cultures on potato-dextrose agar, using either sclerotia or pieces of the advancing margin of the mycelium as inoculum, were typical of the fungus on this medium and produced numerous tan sclerotia about 1 mm. in diameter.

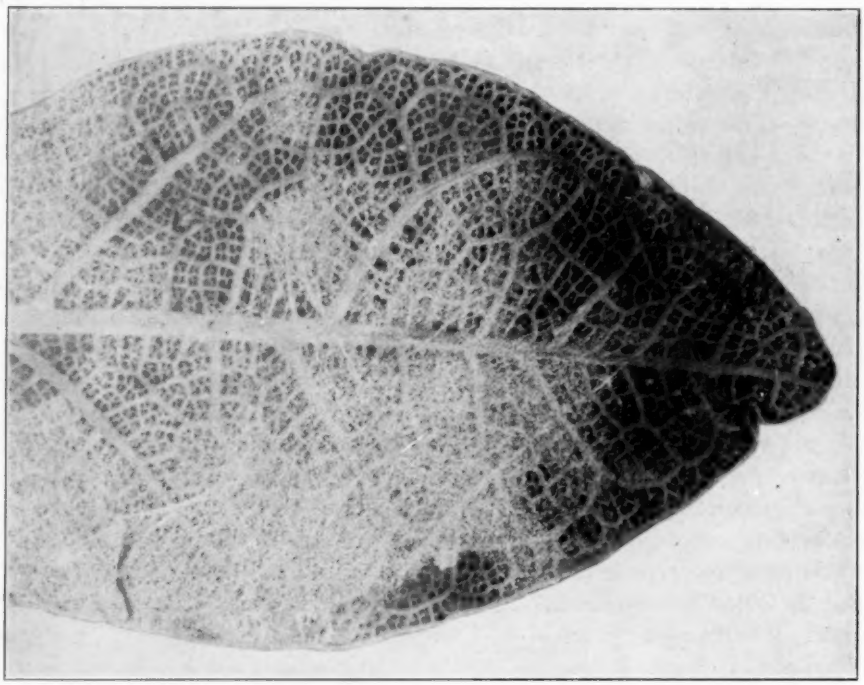

FIG. 1. Hymenium of *Pellicularia rolfsii* on climbing fig leaf. (Mag. × 5.)

While material was being collected for a permanent record of the fungus on this new host, a basidial form was discovered on a few leaves. This was found in considerable quantity later when 2 or 3 sunny days followed prolonged (4- to 6-day) rainy periods. It occurred only on the underside of leaves (Fig. 1) always on the shaded side, always on leaves in the middle or inner layers of vines, and usually several inches behind the actively advancing margin of the fungus. A few patches of this basidial stage occurred in June and July of 1946 on leaves in the same general area.

The hymenium is very coarsely areolate at first, consisting of clusters of basidia arranged in lines on a very tenuous subiculum. As more basidia are formed, the hymenium becomes more dense but never forms a continuous or fleshy layer, and, when fully developed, it is not more than 30 to 40 μ in

thickness. The color of the fruiting hymenium is putty (M. & P. 11–B–2)[1] but in herbarium specimens a year old, it is more gray (M. & P. 42–A–2 or 41–A–2).[1] The basidia are obovoid, 7 to 9 μ long by 4 to 5 μ wide. Each basidium bears 2 or 4 parallel or divergent sterigmata, 2.5 to 4 or occasionally 6 μ long. The spores are elliptical to obovate, rounded above, rounded or pointed at the base, apiculate, 3.5 to 5 μ by 6 to 7 μ, hyaline and smooth. No cystidia or incrusted hyphae have been found.

Two species of *Corticium* have been reported as the basidial stage of *Sclerotium rolfsii* Sacc., namely *Corticium centrifugum* (Lev.) Bres. (reported by Goto[2] and Curzi[3]) and *C. rolfsii* (Sacc.) Curzi (reported by Curzi,[3] Milthorpe,[4] Barrett,[5] and Mundkur[6]). The spore measurements of the *Corticium* on fig fall within the limits given for *C. centrifugum* but such characteristics as color and thickness of the hymenium are quite different. The basidia, sterigmata, and spores agree very well with those described for *C. rolfsii* and since the other characters of that species were described from material on culture media and hence are hardly comparable to naturally produced fructifications, discrepancies here may be disregarded. For the present, the basidial stage on the fig leaves is regarded as *Corticium rolfsii* (Sacc.) Curzi, but inasmuch as Corticium species having an areolate hymenium, short-celled, stout hyphae, right-angled branching of the mycelium and stout basidia have been segregated in the genus *Pellicularia*, the combination *Pellicularia rolfsii* (Sacc.) nov. comb. is proposed as the correct name.

Fifteen single-basidiospore cultures, obtained by suspending fresh hymenia on fig leaves over dishes of potato-dextrose agar, have been compared and considerable variations were noted in color, marking, size, and frequency of the sclerotia and in characteristics of the mycelium growth. Some isolates resembled typical *Sclerotium rolfsii* Sacc., others resembled *S. delphinii* D. S. Welch, and some were intermediate. All isolates are parasitic on *Lupinus angustifolius* L.—ERDMAN WEST, Florida Agricultural Experiment Station, Gainesville, Florida.

Aster Yellows and Its Vector on Potatoes in Nebraska.—In recent years it has been stated[1, 2] that aster yellows is transferable to potatoes by the aster

[1] Maerz, A., and M. Rea Paul. A Dictionary of Color. 1st Ed., 207 + VII pp. McGraw-Hill. New York. 1930.

[2] Goto, K. On the perfect stage of *Sclerotium rolfsii* Sacc. produced on culture media. Preliminary report. Jour. Soc. Trop. Agric. **2**: 165–175. 1930.

[3] Curzi, M. Studi su lo "*Sclerotium rolfsii.*" Boll. R. Staz. Pat. Veg., N. S. **11**: 306–373. 1931.

[4] Milthorpe, F. L. Studies on *Corticium rolfsii* (Sacc.) Curzi (*Sclerotium rolfsii* Sacc.). Proc. Linn. Soc. N. S. W. **66**: 65–75. 1941.

[5] Barrett, J. T. Observations on the basidial stage of *Sclerotium rolfsii*. Phytopath. **24**: 1137–1138. 1934.

[6] Mundkur, B. B. Perfect stage of *Sclerotium rolfsii* in pure culture. Indian Jour. Agric. Sci. **4**: 779–781. 1934.

[1] Leach, J. G. Further experiments on the cause of purple-top wilt of potatoes. Phytopath. **29**: 14. 1939.

[2] Younkin, S. G. Purple-top wilt of potatoes caused by aster-yellows virus. Amer. Pot. Jour. **20**: 177–183. 1943.

leafhopper, *Macrosteles divisus* (Uhl.). Reportedly the disease has become of serious economic concern in certain commercial potato-producing sections of the North Central States. Since both aster yellows and its insect vector occur widely in Nebraska, greenhouse and field tests were undertaken to determine the effect of the disease on potatoes in this State.

During the winters of 1943 and 1944 six inoculation experiments involving 93 potato plants were conducted in the greenhouse at Lincoln. Two- and 4-hill units of Triumph variety potatoes were used. Viruliferous insects were placed on caged potato plants at the time the plants were breaking through the ground. At the end of 15 days the leafhopper-exposed plants were removed from the cage and fumigated. The number of insects used in the different tests varied from 5 to 75, but was usually 25 or more; most of the insects were nymphs at the time of confinement on potatoes. Control plants consisted of related plants which were exposed to nonviruliferous insects, and related plants not subjected to insect feeding. In each of the first four tests healthy aster plants were exposed to 5 viruliferous leafhoppers from the same colony used to inoculate the potatoes. Typical aster yellows developed in the asters. All potato plants were held until maturity, when the tubers were dug and placed in storage. The tubers were indexed in the greenhouse the succeeding fall when all produced healthy-appearing plants.

Of 56 potato plants exposed to viruliferous *Macrosteles divisus*, 2 developed symptoms typical of those described by Younkin[2] and others for aster yellows on potatoes; one other showed symptoms suspiciously like those described but not positively identifiable. The first symptoms appeared 35 to 40 days after inoculation.

During the summer of 1944, one or two plants, 8 to 10 inches high, of each of nine 4-hill tuber units growing in the field at Scottsbluff were caged and exposed to the feeding of 10 viruliferous leafhoppers for 10 days. One plant of the tuber unit in each of 4 cages was exposed to 10 nonviruliferous leafhoppers for a similar period. No symptoms appeared on any of the exposed plants.

Leafhoppers were collected in several commercial potato plantings in the summer of 1944 in Scotts Bluff County, and 5 of these were placed on each of 7 aster plants. Three plants developed typical aster-yellows symptoms.

Field collections of *Macrosteles divisus* were made at 2- to 7-day intervals from July 14 to October 4, 1943, in four potato fields planted on four different dates. In addition, numerous field collections have been made in the same area in connection with other problems from 1940 to 1945, inclusive. These collections indicate that *M. divisus* does not become abundant on potatoes until early August.

Cropping practices and climatic conditions in the commercial potato-growing sections of western Nebraska do not appear to be conducive to the development of aster yellows on potatoes. Although viruliferous leafhoppers were found in potato fields, the time elapsing between the appearance

of *Macrosteles divisus* in significant numbers in potato fields and maturity of the potato plants is usually less than the time required for aster-yellows symptoms to develop after inoculation.—JAMES H. JENSEN, formerly of the Department of Plant Pathology, and H. DOUGLAS TATE, formerly of the Department of Entomology, Nebraska Agricultural Experiment Station, Lincoln, Nebraska.

BOOK REVIEWS

BISBY, G. R. *An Introduction to the Taxonomy and Nomenclature of Fungi.* 117 pp. The Imperial Mycological Institute. Kew, Surrey, England. 1945. 5s or $1.25.

The sixteen chapters in this book are divided, following the introductory chapter, into: Part 1, Taxonomy, and Part 2, Nomenclature. Part 1 is mainly of interest to the beginning student in plant pathology or mycology. After addressing a chapter to the amateur, the writer discusses, in turn, how to choose a fungus group for study, the equipment needed, suggestions on collecting, examining, recording, measuring, and culturing fungi; naming and describing them; preserving material; and finally, suggestions on publishing and illustrating.

Part 2, which is considerably more technical than Part 1, has chapters on the categories of fungi, synonymy, types and the type method, diagnoses, rules of nomenclature, literature citations, and index. The value of this part of the book is greatly increased by the inclusion of the International Rules of Botanical Nomenclature, together with notes and examples of fungus names. As many plant pathologists have not had ready access to these rules, this feature alone is sufficient to recommend this inexpensive little book to the plant pathologist who works with fungi.—RODERICK SPRAGUE, Washington State College, Pullman, Washington.

DUBOS, RENÉ J. *The Bacterial Cell in Its Relation to Problems of Virulence, Immunity, and Chemotherapy.* 460 pp., 53 figs. Harvard University Press, Cambridge, Mass. 1945. $5.00.

This excellent book is Harvard University Monograph in Medicine and Public Health Number 6. Thus it deals almost exclusively with human pathogens. It covers the following general topics: (1) materials, problems, and methods; (2) cytology; (3) physicochemical and staining properties; (4) analysis of cellular structure by biochemical and biological methods; (5) variability; (6) nature of virulence; (7) immunization; (8) bacteriostatic and bactericidal agents; (9) trends and perspectives; and (Addendum, by C. F. Robinow) nuclear apparatus and cell structure of rod-shaped bacteria.

This book is really important for those interested in bacteria. However, it apparently contains only casual or no mention of numerous important items concerned with bacteria pathogenic to farm animals, with plant pathogens (4 references), with legume root-nodule bacteria (2 references), and with forms important in agriculture and industry.

The chapter on virulence is particularly noteworthy for the plant pathologist (but there is no mention of plant pathogens). Descriptions appear of (1) host-parasite relationships, (2) resistance of the parasite to the defense mechanisms, (3) factors affecting invasiveness, (4) toxic action, and (5) independent variation of the different components of virulence. The author emphasizes that virulence is not a single character but a combination of a number of different characters. These may vary independently. This neglected viewpoint deserves continued emphasis for those studying critical questions of virulence. It is complementary to questions of disease resistance.—A. J. RIKER, University of Wisconsin, Madison, Wisconsin.

GILMAN, JOSEPH C. *A Manual of Soil Fungi.* ix + 392 pp. 135 figs. The Collegiate Press, Inc., Ames, Iowa. 1945. $5.00.

In his preface Dr. Gilman states that this book is an expanded revision of the paper "A Summary of the Soil Fungi" published by Dr. E. V. Abbott and himself in 1927. The deserved popularity of that earlier contribution is a good index to the potential value of this present work. The compilation covers principally those soil fungi that have been isolated and grown on artificial media and excludes some special groups of fungi such as soil-borne plant pathogens not isolated directly from the soil. The often tedious task of identifying the fungi associated with soil is made easier by the many dichotomous keys, line drawings and adequate descriptions. Plant pathologists should welcome this book, for it brings into one volume descriptions of many of those fungi that occur not only in the soil but on seeds and other plant parts.—IAN W. TERVET, University of Nebraska, Lincoln, Neb.